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Official Organ of
The American Phytopathological Society

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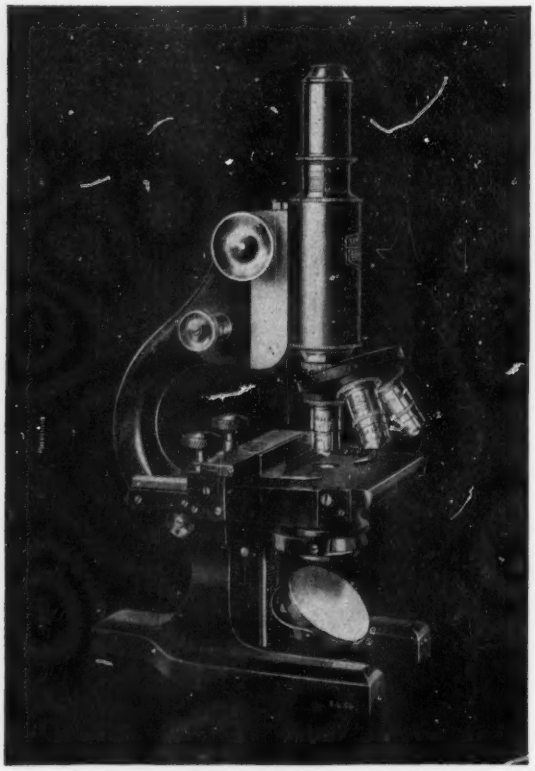

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An International Journal
Official Organ of The American Phytopathological Society

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WARNER JACKSON MORSE

PHYTOPATHOLOGY

VOLUME 21 NUMBER 12

DECEMBER, 1931

WARNER JACKSON MORSE
1872–1931

EDITH M. PATCH

Warner Jackson Morse came to the Maine Agricultural Experiment Station in 1906 to head the Department of Plant Pathology. In 1921 he became Director of the Experiment Station and he still held this title at the time of his death on the twenty-fifth of March, 1931.

During his quarter-of-a-century residence in Maine, I knew him as colleague, friend, and neighbor and, for the last decade, as an official superior. Because of this long acquaintance, I have been asked to write this sketch.

To speak appropriately of Doctor Morse is to speak informally and with simplicity. He drove one day, a few years ago, into the country cemetery across the river from his home. His glance sought the hills in the far distance, and he said to his wife, "This is good enough for me." It was fitting that his words were remembered and that, when the time came for them, his last rites were observed in the town of Orono, which had been his dwelling place for about twenty-five years.

Perhaps lack of ostentation is natural to the country bred. In keeping, certainly, with the associations of his early life, were his later occupations. This man was always at heart a farmer. The products of the soil interested him. The subjects that he taught, as a young man, were botany and bacteriology. Later, his research work was concerned with the diseases of plants to the end that the health of certain crops might be safeguarded. Obviously, his earnest interest in agricultural matters, from practical as well as from experimental aspects, contributed significantly to his economic service to Maine.

Outdoor interests were the background also of his recreational pursuits. He found keen pleasure in working in his vegetable and flower gardens. He made a squirrel welcome to picnic luncheons on the ledge of his office window. Bluebirds and tree swallows nested in boxes he made and placed for them on the trees about his home. He planted bushes that grosbeaks might enjoy their fruit. And, of all the books that he read during his long illness, perhaps none gave him greater enjoyment than "Jack Miner and the Birds."

Although interest and industry are necessary to the sort of success achieved by Doctor Morse, they are not in themselves sufficient. Tenacity of purpose, too, is a requisite; and this he had.

And, since he headed the Maine Agricultural Experiment Station during those most meager years immediately preceding the Purnell appropriations, his conservativeness stood the institution in good stead. Indeed conservatism might well be indicated as one of his outstanding qualities. The known and the tried made a much stronger appeal to him than the new and the untested. He never forgot an old friend. An interest account in a stable savings bank outweighed any temptations to invest in stocks and bonds. And these same tendencies, characteristic of his personal affairs, influenced his official attitude.

Thus, under his direction, new departments, new lines of research, new projects were ventured upon; but only when he became convinced of their importance and significance.

His conservatism, however, could not be interpreted as a concession in quality. For it was only the best that appealed to him—whether in the choice of tools and mechanical devices for his personal use or in sanctioning standards and ideals in his professional and official capacities.

In keeping with his other sturdy qualities was his unvarying honesty and indiscriminating justness. No one associated with him as colleague or subordinate had reason to feel unfairly treated. There was no department or investigator favored at the expense of another.

Warner Jackson Morse was born in Waterbury Center, Vermont, October 30, 1872. He was the son of Daniel Jackson and Jane (McKee) Morse. In 1898 he married Mary Leland, of Johnson, Vermont, who survives him, as does their daughter, Ruth Morse Burbank. Surviving him also are two sisters, Mrs. Ida Grout, of Waterbury, Vermont, and Mrs. Mae Heath, of Chautauqua, New York.

He was graduated from the Johnson (Vermont) Normal School in 1893. From the University of Vermont he received the earned degrees B.S. in 1898 and M.S. in 1903. The same institution bestowed upon him the honorary degree Sc.D. in 1923. He pursued his doctorate work at the University of Wisconsin, where he took his Ph.D. in 1912.

He was a teacher of natural sciences, Montpelier (Vermont) Seminary, 1899–1901. At the University of Vermont he was instructor of botany, 1901–1905, and assistant professor in bacteriology, 1905–1906. He was also assistant botanist at the Vermont Agricultural Experiment Station from 1901 until 1906, when he came to the sister institution in Maine.

He was a member of Sigma Xi, Phi Kappa Phi, Kappa Sigma, Alpha Zeta, The American Phytopathological Society (charter member), and the

Botanical Society of America, and fellow of the American Association for the Advancement of Science. He was a Mason.

His published papers total more than fifty titles.

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FURTHER STUDIES ON THE FUNGICIDAL EFFICIENCY OF CHEMICAL DUSTS CONTAINING FURFURAL DERIVATIVES

J. J. WILSON[1] AND C. S. REDDY[2]

INTRODUCTION

Organic-mercury compounds having a furan ring were found by the junior writer,[3] in 1927, to be applicable as dust fungicides for the control of the dry-rot seedling blights of corn. The favorable preliminary results led to a more intensive study to determine what methods of preparation and what concentrations of the organic-mercury compounds were most beneficial in the control of three seed-borne diseases of corn. Laboratory and field experiments were conducted in 1928 and 1929 for these purposes and to determine further the merits of certain laboratory biological tests by correlating laboratory and field results.

COMPOUNDS STUDIED

The structural formulae of these organic-mercury compounds having the furan ring are not yet fully understood. Their methods of preparation have been described in a previous paper by the junior writer.[4] For convenience in subsequent discussion they are divided into three groups.

(A) E2C. The reaction product formed as precipitate when a solution of mercuric chloride is stirred into a furfural solution previously subjected to the Cannizzaro reaction and to which ammonia has been added.

(B) E4. The reaction product formed as a precipitate when a solution of mercuric nitrate is stirred into a furfural solution previously subjected to the Cannizzaro reaction.

(C) G1. The reaction product formed as a precipitate when solutions of mercuric chloride and furfuramide are stirred together. Either alcohol, acetone, or aqueous solution of ammonium chloride is used as the solvent for the mercuric chloride and furfuramide.

The resultant precipitate of each of the three groups was dried, finely ground, and mixed with talc as a filler. In this manner a number of fungi-

[1] Research Fellow, Crop Protection Institute, 1928, on utilization of furfural products as seed disinfectants, stationed at the Plant Pathological Laboratories, under the direction of Dr. I. E. Melhus, Iowa State College, Ames, Iowa.

[2] Assistant Chief of Botany and Plant Pathology Section, Iowa State College, Ames, Iowa.

[3] Reddy, C. S. Fungicidal efficiency of chemical dusts containing furfural derivatives. Phytopath. 20: 147–168. 1930.

[4] *Loc. cit.*

cidal dusts having different concentrations of the toxic agents were prepared. These experimental dusts are designated by numbers and the nature of the organic-mercury precipitate may be identified by the symbol E2C, G1, or E4.

In order to find out whether or not these fungicides could be manufactured without difficulty or excessive cost, all of the E2C and G1 dusts, used in 1928, were prepared by the Miner Laboratories, Chicago, Illinois.

LABORATORY EXPERIMENTS—1928

The number of fungicidal dusts prepared was much larger than could be given field experimentation and had to be reduced on the basis of data secured from laboratory experiments. The importance of these preliminary tests was plainly evident. Unless there was a high correlation between the laboratory and field results, the most beneficial dust fungicide might be discarded without field trial. Previous experiments had shown that the visible-root sand-culture method devised by Raleigh[5, 6] had merit in selecting dusts that combined high fungicidal value with little or no injurious effect upon living corn-plant tissues. This method was used in these experiments and later was varied to overcome some of the difficulties of its manipulation. The variation consisted in the use of dairy cups in place of the crystallization dishes. This permitted any slight excess of water to drain through holes punched in the bottom of the cups. Also, when dairy cups were used the seed was placed midway rather than on the bottom, where it was thought soluble toxic agents might accumulate. Wherever this preliminary method is used, it is designated the dairy-cup sand-culture method.

A detailed study of the E2C, G1, and E4 compounds required that variations in the method of preparation and in the amount of the organic-mercury precipitates be made and tested.

One hundred and forty kernel lots of a standard variety of dent corn known to be highly infected with *Diplodia zeae* (Schw.) Lev. were treated with each of 12 E2C, 43 G1, and 2 E4 dusts. Seven replications of 20 kernels of each seed-treatment lot were planted in crystallization dishes. These dishes were 10 cm. in diameter and 5 cm. deep. Washed river sand was used to cover the corn to a depth of 4 cm. The cultures were held at 18° to 22° C. and the soil moisture was maintained near 75 per cent of saturation.

After 12 days' growth, the plants were washed free from sand and weighed, and the growing plants and lesions caused by Diplodia were

[5] Raleigh, W. P. A preliminary method of measuring the relative efficiency of seed corn disinfectants. (Abst.) Phytopath. 18: 140. 1928.

[6] Infection studies of *Diplodia zeae* (Schw.) Lev. and control of seedling blights of corn. Iowa Agr. Exp. Sta. Res. Bul. 124. 1930.

TABLE 1.—*Laboratory experiments on the fungicidal action of different concentrations of E2C dusts on seed corn infected with Diplodia zeae, Ames, Iowa, 1928*

Dust No.	Constituent parts			Temperature precipitate dried	Growing plants	Green weight	Relative value	Plants with lesions
	Furfural	Mercuric chloride	Talc					
				°C.	*Number*	*Grams*		*Per cent*
1	5	6.6	50	20 to 24	110	171	266	12.7
2	"	"	60	"	120	186	289	25.8
3	"	"	75	"	124	196	310	18.5
4	"	"	100	"	125	194	301	18.4
5	"	"	150	"	123	189	290	21.9
6	"	"	300	"	121	196	317	16.5
7	"	"	50	90	127	198	309	4.7
8	"	"	60	"	131	206	324	9.9
9	"	"	75	"	131	195	290	3.0
10	"	"	100	"	127	179	252	12.5
11	"	"	150	"	127	203	324	6.2
12	"	"	300	"	127	199	312	9.4
Check[a] (not treated)					120	172	247	44.2

[a] Computed from 27 replications.

counted in each treatment and check. From these data the average green weight per plant and the percentage of plants with lesions were computed for each seed treatment. An arbitrary method of expressing the relative value of each treatment was obtained by squaring the total green weight and dividing by the number of growing plants. In this way the value was determined by a combination of individual and total plant weights.

The interpretation of the biological response in such laboratory tests involves a study of the fungicidal effectiveness of the dust and the physiological action of the treatment on the growth of the corn seedling. Care was taken in the study of each individual dust to determine whether or not depressing effects were produced on plant growth. In measuring the fungicidal efficiency of the dusts, which were selected to be used in the field trials, low lesion counts were not considered sufficient, but were used in conjunction with green weight and number of living plants.

Table 1 shows the results obtained in laboratory trials with 12 E2C dusts. The treated seeds produced in every instance a lower percentage of plants with lesions and a higher relative value for total green weight than seed not treated. The data also show that the plants from seeds treated with E2C dusts dried at 20° to 24° C. had higher lesion counts than those treated with E2C dusts dried at 90° C. This probably was due to the more resinous nature of the E2C precipitate when dried at the lower temperature. Resinous precipitates are often difficult to grind and mix with talc and such difficulties may cause dust fungicides to have poor physical properties, resulting in a reduction of fungicidal efficiency. E2C Nos. 8 and 11 were selected for field trials. These 2 dusts had equal relative values and showed low percentages of plants with lesions.

Five different G1 precipitates were made by varying the proportions of furfuramide and mercuric chloride and by using different solvents. From these precipitates 43 different dust fungicides were made by varying the amount of talc filler.

Laboratory experimental results of these 43 G1 dusts, as shown in tables 2 and 3, indicate that the variation in the quantity of furfuramide or mercuric chloride produced no marked effect. There were, however, differences in the resinous character of the resultant precipitates, which had to be taken into consideration in the final selection of the dusts. The lower concentrations of the G1 precipitate gave poorer control of the disease and, therefore, were associated with higher lesion counts. Without exception, all concentrations of the G1 precipitate had some fungicidal value because the percentage of plants with disease lesions was lower and the relative value higher in all of the treatments than in the checks not treated.

On the basis of the data presented in tables 2 and 3, G1 dusts Nos. 13, 50, 51, 53, and 61 were selected for field trial. Number 13 contained

TABLE 2.—*Laboratory experiments on the fungicidal action of different concentrations of G1 dust, on seed corn infected with Diplodia zeae, Ames, Iowa, 1928*

TABLE 2.—*Laboratory experiments on the fungicidal action of different concentrations of G1 dust, on seed corn infected with Diplodia zeae, Ames, Iowa, 1928*

Dust No.	Constituent parts			Temperature precipitate dried	Growing plants	Green weight	Relative value	Plants with lesions
	Furfuramide	Mercuric chloride	Talc					
				°C.	*Number*	*Grams*		*Per cent*
13	5	6	50	20 to 24	125	206	339	0.0
14	"	"	60	"	130	203	317	0.7
15	"	"	75	"	127	199	312	6.2
16	"	"	100	"	126	197	308	6.3
17	"	"	150	"	125	196	307	6.4
18	"	"	50	90	129	206	329	2.3
19	"	"	60	"	121	195	314	4.1
20	"	"	75	"	130	202	314	1.5
21	"	"	100	"	127	196	304	14.1
22	"	"	150	"	134	198	316	4.0
23	7	3.5	50	20 to 24	129	206	329	3.1
24	"	"	60	"	130	188	272	6.9
25	"	"	75	"	121	187	289	8.2
26	"	"	100	"	132	203	315	8.3
27	"	"	150	"	134	206	317	18.6
28	"	"	50	90	126	192	292	3.9
29	"	"	60	"	122	183	274	5.7
30	"	"	75	"	130	210	339	5.3
31	"	"	100	"	117	187	299	17.9
32	"	"	150	"	121	181	271	20.6
33	3.5	7	50	20 to 24	122	193	308	9.8
34	"	"	60	"	131	207	317	3.8
35	"	"	75	"	125	205	336	9.6
36	"	"	100	"	124	203	334	3.2
37	"	"	150	"	123	189	290	9.7
38	"	"	50	90	120	191	304	2.5
39	"	"	60	"	129	202	314	3.1
40	"	"	75	"	124	194	303	3.2
41	"	"	100	"	125	210	353	11.2
42	"	"	150	"	132	197	295	12.8
61	5	15	180	"	122	201	331	4.9
Check[a] (not treated)					120	172	247	44.2

[a] Computed from 27 replications.

TABLE 3.—*Laboratory experiments on the fungicidal action of different concentrations of G1 dusts (ammonium chloride solvent) used on seed corn infected with Diplodia zeae, Ames, Iowa, 1928*

Dust No.	Constituent parts					Temperature precipitate dried	Growing plants	Green weight	Relative value	Plants with lesions
	Furfuramide	Mercuric chloride	Water	Ammonium chloride	Talc					
						°C.	*Number*	*Grams*		*Per cent*
43	15	5	500	30	50	20 to 24	127	195	299	7.8
44	"	"	"	"	100	"	127	196	302	2.3
45	"	"	"	"	150	"	126	188	280	11.9
46	"	"	"	"	200	"	126	202	325	7.1
47	"	"	"	"	250	"	127	210	347	11.8
48	"	"	"	"	300	"	132	211	337	13.6
49	"	"	"	"	50	90	122	189	293	2.4
50	"	"	"	"	100	"	132	218	360	3.7
51	"	"	"	"	150	"	130	208	333	7.6
52	"	"	"	"	200	"	132	199	300	6.8
53	"	"	"	"	250	"	122	197	317	10.6
54	"	"	"	"	300	"	199	190	303	24.3
Check[a] (not treated)							120	172	247	44.2

[a] Computed from 27 replications.

TABLE 4.—*Summary of laboratory data on the 14 fungicidal dusts selected for field experiments. Ames, Iowa, 1928*

Dust No.	Identification symbol or name	Constituent parts					Solvents other than water	Relative value	Comparison with highest relative value
		Furfural	Furfuramide	Mercuric chloride	Mercuric nitrate	Talc			
									Per cent
8	E2C	5		6.6		60	none	324	90
11	E2C	5		6.6		150	none	324	90
13	G1		5	6.0		50	$(CH_3)_2CO$	340	94
50	G1		15	5.0		100	NH_4CL	360	100
51	G1		15	5.0		150	NH_4CL	333	92
53	G1		15	5.0		250	NH_4CL	316	87
58	E4	30			5	95	none	359	99
61	G1		5	15.0		95	$(CH_3)_2CO$	331	91
61a	G1		5	15.0		93	$(CH_3)_2CO$		
61b	G1		5	15.0		97	$(CH_3)_2CO$		
67	F–13							317	88
63	Bayer dust							301	83
64	Semesan Jr.							328	91
65	Merko[a]							269	74
Check (not treated)								247	69

[a] The low relative value of Merko in comparison with the other commercial dusts is probably due to overwatering two cultures in the Merko series.

approximately the same concentration of toxic agent as the G1 dust used in 1927. Numbers 50, 51, and 53 represent different concentrations of G1 prepared by using ammonium chloride as the solvent. Number 61 was chosen because the precipitate was not resinous and could be easily ground and mixed with a talc filler. Three different concentrations of No. 61 were made up in large quantities for field trials. The concentrations were made to approximate as closely as possible dusts Nos. 30, 36, and 41. These last 3 dusts had high relative values but were not easily prepared because of difficulty in grinding the precipitate.

The laboratory data on 14 fungicidal dusts selected for 1928 field experimental study are summarized in table 4.

LABORATORY EXPERIMENTS—1929

In 1929 a study was made of the relative fungicidal efficiency of G1 in concentrations of 3, 4, and 5 per cent of the toxic agent, mercury furfuramide. G1 dusts of these three strengths were compared with Merko in

TABLE 5.—*Laboratory data on emergence, disease lesions, and relative-weight value obtained by treating 2 lots of Diplodia-infected seed corn with dust Nos. 61, 61b, Sterocide, and Merko. Ames, Iowa, 1929*

Dust No.	1928 Diplodia-infected seed				
	Growing plants	Green plant weight	Relative value	Comparison with highest relative value	Plants with lesions
	Number	*grams*		*Per cent*	*Per cent*
(G1–5%) 61	79	165	344	100	13.3
(G1–3%) 61b	77	162	341	99	15.0
(G1–4%)[a]	78	159	324	94	12.2
Merko	78	160	328	95	8.0
Check (not treated)	65	125	238	69	35.0
	1927 Diplodia-infected seed				
(G1–5%) 61	90	179	356	92	1.1
(G1–3%) 61b	87	175	352	91	4.5
(G1–4%)[a]	88	176	552	91	2.0
Merko	87	184	385	100	0.0
Check (not treated)	86	159.5	294	76	13.8

[a] Sterocide Average of 9 lots.

laboratory trials on 2 different lots of Diplodia-infected seed corn. The dairy-cup sand-culture method was used in making the laboratory biologic tests. Six replications of 15 kernels were planted from both seed lots after being treated by the 4 dusts. The 3 and 5 per cent concentrations of G1 were dusts Nos. 61 and 61b of the 1928 experimental series. The 4 per cent concentration of G1 is the commercial form placed on the market in 1929 and known by the trade name, Sterocide. The laboratory data from 9 individual lots of Sterocide (4 per cent toxic agent) were averaged, and these data are compared in table 5 with data from dust No. 61 (5 per cent toxic agent), No. 61b (3 per cent toxic agent), and Merko.

All 4 dust seed treatments (data in table 5) produced a decided increase in the weight of the plants and a reduction in the percentage of plants with lesions. Seed treated with dust No. 61b (3 per cent toxic agent) produced a higher percentage of plants with lesions than seed treated with No. 61 (5 per cent toxic agent) or Sterocide (4 per cent toxic agent). The data indicated that 61b was possibly a little too weak in fungicidal action.

FIELD EXPERIMENTS

In order to determine the reliability of the laboratory methods used in measuring the fungicidal efficiency of seed-corn dust fungicides, field trials were conducted with the dusts selected in the 1928 and 1929 laboratory experiments.

Field experiments—1928. The 10 best E2C, G1, and E4 dusts selected in laboratory trials and 3 commercial dusts (summary of results in table 4) were used in seed-treatment experiments on 2 different lots of diseased seed corn and a lot of nearly disease-free seed corn. All 3 lots of seed corn were selected by an individual ear sand-tray-germinator test of a high-yielding local strain of Reids Yellow Dent Corn. One of the 2 lots of diseased seed corn was infected with *Diplodia zeae* and was selected from ears showing large infected zones with viable kernels. The other lot of diseased seed was infected with *Basisporium gallarum* Moll. and was selected from ears having viable kernels with adhering Basisporium spores. The nearly disease-free seed was secured from sound ears that were germinator-selected to eliminate practically all diseased kernels.

The diseased seed corn was used to measure the relative fungicidal efficiency of the different dusts and the nearly disease-free seed corn was used to detect any injurious effect caused by the different dusts on the growth of the plants. Field stand and acre yield were used to measure these two factors.

Thirty hill plots, replicated 10 times, were used for testing each dust treatment and repeated for each of the 3 different seed lots. Seed not treated was included as checks in each replication. The rate of planting

was 2 kernels per hill in all plots except those planted with Basisporium-infected seed in which the rate was 3 kernels per hill. The kernels for each hill were counted and planted by hand.

The plots were planted May 5 in order to subject the seed to any possible cold weather and unfavorable growing conditions that might accompany a moderately early planting. This aim, however, was not realized because the season of 1928 proved unusually mild in temperature. The seed germinated quickly and the plants made a rapid, continued growth. The seed-treatment plots were cultivated throughout the season in the same manner as a regular field of corn. After the first and second cultivation, all plants covered by the plow were uncovered to prevent a mechanical loss in stand.

The effect of different dusts on stand. The field-stand data presented in table 6 were taken 3 weeks after planting. On nearly disease-free seed,

TABLE 6.—*Field stands from nearly disease-free, Basisporium-infected, and Diplodia-infected seed corn not treated and treated with 10 dusts containing organic mercury with a furan ring and three commercial dusts. Ames, Iowa, 1928*

Dust No.	Nearly disease-free seed		Basisporium-infected seed		Diplodia-infected seed	
	Stand	Increase or decrease	Stand	Increase or decrease	Stand	Increase or decrease
	Per cent	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*
Check (not treated)	91.67		70.67		62.67	
8 (E2C)	89.26	−2.63	70.00	− .73	68.00	+ 5.81
11 (E2C)	92.04	+ .40	71.89	+1.33	70.84	+ 8.90
13 (G1)	87.50	−4.55	72.34	+1.82	76.00	+14.53
61a (G1)	90.39	−1.40	71.67	+1.09	72.84	+11.80
61 (G1)	91.17	− .54	74.45	+4.12	69.84	+ 7.81
61b (G1)	89.84	−1.99	73.23	+2.79	74.00	+12.35
65 (Merko)	91.67	−1.63	70.34	− .36	76.00	+14.53
67 (F13)	90.34	−1.45	74.78	+4.48	70.50	+ 8.54
50 (G13)	90.34	−1.45	74.89	+4.60	76.50	+15.75
51 (G13)	92.17	+ .54	75.56	+5.33	70.50	+ 8.54
53 (G13)	90.50	−1.28	73.56	+3.15	73.67	+11.99
58 (E4)	91.34	− .37	74.12	+3.76	74.84	+13.26
63 (Semesan Jr.)	91.00	− .73	73.34	+2.91	73.67	+11.99
64 (Bayer Dust)	91.00	− .73	71.56	+ .97	70.34	+ 8.36

2 treatments, Nos. 8 and 13, reduced the stand sufficiently to indicate injury. Both dusts contained a high concentration of the active precipitate. Dust No. 8 reduced the stand 2.63 per cent, and No. 13, 4.55 per cent. Percentage increases or decreases in the other 12 dust treatments were relatively small. Dust treatments of Basisporium-infected seed with Nos. 67, 50, and 51

increased the stand more than 4 per cent and treatments with Nos. 61 and 58 were nearly as efficient in increasing the stand.

Significant increases in stand were secured by treatment of Diplodia-infected seed corn. The treated seed produced 5.81 to 15.75 per cent more plants than the checks (not treated).

Under these favorable conditions for corn growth, such increases in the percentage of stand from seed treatment are secured only on badly diseased seed. Continued unfavorable weather, at the beginning of seed germination, usually results in much larger increases in stands.

Yields obtained. The plots were harvested during the last week of October. The corn was exceptionally well matured and dry. Acre yields of ear corn (75 lbs. per bu.) from the 3 different seed lots were computed for 10 replications of each treatment and check.

Table 7 shows that treatments of nearly disease-free seed corn produced

TABLE 7.—*Acre yields of field plots planted with nearly disease-free seed corn treated with 10 dusts having E2C, G1, and E4 as their toxic agents and 4 commercial dusts. Ames, Iowa, 1928*

Dust No.	Acre yield		Increase or decrease	Odds[a]
	Not treated	Treated		
			Bushels	
8 (E2C)	69.2	69.8	+0.6	1:1
11 (E2C)	69.2	70.4	+1.2	1:1
13 (G1)	69.2	68.8	−0.4	1:1
61a (G1)	68.4	70.7	+2.3	6:1
61 (G1)	68.4	71.6	+3.2	49:1
61b (G1)	68.4	71.0	+2.6	6:1
65 (Merko)	67.7	68.1	+0.4	1:1
67 (F13)	67.7	71.0	+3.3	252:1
50 (G1)	67.7	70.6	+2.9	11:1
51 (G1)	67.7	70.0	+2.3	29:1
53 (G1)	68.8	69.0	+0.2	1:1
58 (E4)	68.8	70.5	+1.7	4:1
63 (Semesan Jr.)	68.8	72.4	+3.6	171:1
64 (Bayer Dust)	70.1	71.1	+1.0	1:1

[a] Student's method.

no outstanding increases in yield. Dusts Nos. 67, 63, and 61 increased the yield slightly over 3 bushels and the odds were significant. Only 1 dust, No. 13, produced any decrease in yield and this was not significant. (Odds 1:1.) These data indicate that no injury has been caused by seed treatment.

The yield data from the field plot planted with Basisporium-infected seed are presented in table 8. It will be seen that no significant odds were

TABLE 8.—*Acre yields of field plots planted with Basisporium-infected seed corn treated with 10 dusts having E2C, G1, and E4 as their toxic agents and 4 commercial dusts. Ames, Iowa, 1928*

Dust No.	Acre yield		Increase or decrease	Odds[a]
	Not treated	Treated		
			Bushels	
8 (E2C)	79.6	74.1	−5.5	9:1
11 (E2C)	79.6	78.2	−1.4	2:1
13 (G1)	78.3	79.4	+1.1	2:1
61a (G1)	78.3	79.6	+1.3	2:1
61 (G1)	78.3	80.2	+1.9	5:1
61b (G1)	77.1	81.6	+4.5	11:1
65 (Merko)	77.1	79.0	+1.9	4:1
67 (F13)	77.1	78.5	+1.4	2:1
50 (G1)	77.1	79.4	+2.3	5:1
51 (G1)	79.5	82.9	+3.4	6:1
53 (G1)	79.5	79.7	+0.2	1:1
58 (E4)	79.5	81.5	+2.0	8:1
63 (Semesan Jr.)	81.6	78.0	−3.6	15:1
64 (Bayer Dust)	81.6	82.0	+0.4	1:1

[a] Student's method.

secured in the increase in yield from seed treatment of Basisporium-infected seed corn. The greatest increases in yield were secured with dusts Nos. 61b and 51. Both dusts contained a low percentage of organic mercury. Dusts Nos. 8, 11, and 63, each of which contained a high percentage of organic mercury, decreased the yields.

The yield from the field plot planted with Diplodia-infected seed are presented in table 9 in which it is shown that outstanding increases in yield resulted from treatments of Diplodia-infected seed corn. Dust No. 65 increased the yield 15.1 bu. per acre. It was nearly equaled by dust No. 50, which increased the yield 14.4 bu. per acre. Dusts Nos 65 and 50 gave odds of 4,999 to 1. No. 61 increased the yield only 2.5 bushels, but 61a and 61b increased yields more than 11 bu. per acre. This seems to show that the relative rank of No. 61 was below its deserved position.

These results indicate that a fungicidal efficiency of practical value is present in the dusts containing E2C, G1, and E4 precipitates.

In order to compare the dusts containing the furan ring with the other dusts (3 of the 4 being commercial dusts), the dust treatments were ranked on the basis of increases in acre yield in each of the 3 field plots. It was found that seed treatments containing the furan ring occurred twice in the 4 highest in 1 experiment (Table 7), 4 times out of 4 in another (Table 8), and 3 out of the 4 times in the other field experiment (Table 9).

TABLE 9.—*Acre yields of field plots planted with Diplodia-infected seed corn treated with 10 dusts having E2C, G1, and E4 as their toxic agents and 4 commercial dusts. Ames, Iowa, 1928*

Dust No.	Acre yield		Increase or decrease	Odds[a]
	Not treated	Treated		
			Bushels	
8 (E2C)	52.6	57.8	+ 5.2	15:1
11 (E2C)	52.6	65.0	+12.4	29:1
13 (G1)	54.6	66.3	+11.7	1110:1
61a (G1)	54.6	66.0	+11.4	49:1
61 (G1)	54.6	57.1	+ 2.5	4:1
61b (G1)	52.0	63.7	+11.7	216:1
65 (Merko)	52.0	67.1	+15.1	4999:1
67 (F13)	52.0	61.9	+ 9.9	216:1
50 (G1)	52.0	66.4	+14.4	4999:1
51 (G1)	54.5	62.0	+ 7.5	61:1
53 (G1)	54.5	63.3	+ 8.8	262:1
58 (E4)	54.5	64.2	+ 9.7	100:1
63 (Semesan Jr.)	57.1	67.6	+ 9.5	54:1
64 (Bayer Dust)	57.1	61.4	+ 4.3	5:1

[a] Student's method.

Field experiments—1929. The same concentrations of G1 studied in the laboratory experiments, namely, 3, 4, and 5 per cent of mercury furfuramide, were tested for fungicidal efficiency in field plots. One lot of seed corn infected with *Gibberella saubinetti* (Mont.) Sacc. and the other infected with *Diplodia zeae* were used in seed-treatment experiments with dusts Nos. 61 (G1–5 per cent), 61b (G1–3 per cent), Sterocide (G1–4 per cent), and Merko.

Six replications of 30-hill plots were planted on May 4 with seed corn treated with each dust. The method of planting was similar to that used in the 1928 field trials except that all plots were planted 2 kernels per hill.

Effect on stand. The season was less favorable for corn growth than in 1928, as was shown by the lower percentages of living plants in the 1929 plots.

The field stands following seed treatments with dusts Nos. 61, 61b, Sterocide, and Merko upon Diplodia-infected seed corn and Gibberella-infected seed corn are presented in table 10.

Diplodia-infected seed corn treated with dust No. 61b (G1–3 per cent) has the lowest percentage of stand. The plants in the laboratory trials treated with this same concentration of G1 also had the highest number of lesions. In the same experiments dust No. 61 (G1–5 per cent) consistently

TABLE 10.—*Field stands from Diplodia-infected and Gibberella-infected seed corn treated with dust Nos. 61, 61b, Sterocide, and Merko, Ames, Iowa, 1929*

Dust No.	Diplodia-infected seed		Gibberella-infected seed	
	Stand	Increase	Stand	Increase
	Per cent	*Per cent*	*Per cent*	*Per cent*
(G1–5%) 61	55.7	52.2	82.5	20.4
(G1–3%) 61b	49.8	36.1	74.3	8.5
Sterocide (G1–4%)	50.3	37.4	81.6	19.1
Merko	55.5	51.7	74.0	8.0
Check (not treated)	36.6		68.5	

increased field stands from both Diplodia and Gibberella-infected seed more than 61b (G1–3 per cent) or Sterocide (G1–4 per cent).

This would indicate that the 5 per cent concentration is more efficient as a fungicide than either of the 2 lower concentrations.

Yields obtained. As compared with nontreated seed, treatments with dust No. 61 (G1–5 per cent) and Merko increased the yields from Diplodia-infected seed corn more than 50 per cent (Table 11). Treatments with

TABLE 11.—*Acre yields obtained by treating Diplodia-infected and Gibberella-infected seed corn with dusts Nos. 61, 61b, Sterocide, and Merko, Ames, Iowa, 1929*

Dust No.	Diplodia-infected seed			Gibberella-infected seed		
	Acre yield	Increase	Percentage	Acre yield	Increase	Percentage
	Bushels	*Bushels*		*Bushels*	*Bushels*	
(G1–5%) 61	60.6	21.0	53.0	86.8	11.5	15.2
(G1–3%) 61b	54.5	14.9	37.6	85.0	9.7	12.9
(G1–4%) Sterocide	54.7	15.1	38.1	84.3	9.1	12.1
Merko	61.0	21.4	54.0	84.3	9.0	12.0
Check (not treated)	39.6			75.2		

dust No. 61b (G1–3 per cent) and Sterocide (G1–4 per cent) increased the yields 37.6 and 38.1 per cent, respectively.

A 15.3 per cent increase in yield was secured when Gibberella-infected seed corn was treated with dust No. 61 (G–5 per cent). The effects of the other 3 dusts were much alike, all producing about equal increases in yield.

SUMMARY

The scope of these investigations embraced the synthesis of a new toxic agent, the determination of the proportion of toxic agent to inert matter in the making of a fungicidal dust for seed-corn treatments, and the adjustment of the laboratory methods to commercial manufacture of the product.

A high correlation was found between the expressed fungicidal efficiency measured by laboratory biological tests and the stand and yield per acre secured by field experiments, using the same dust treatments.

Results of these experiments show that laboratory biological tests have been perfected that can be used to measure the relative fungicidal efficiency of seed treatments for corn.

From among 58 different concentrations of the organic-mercury compounds containing a furan ring, a concentration of 5 parts of G1 and 95 parts of talc was found to be the most consistent in giving the best fungicidal action on certain pathogenic organisms, such as *Diplodia zeae, Basisporium gallarum,* and *Gibberella saubinetii* without injurious effects on nearly disease-free seed.

When the fungicidal dusts were ranked on the basis of acre-yield increases in each of 3 large field experiments in 1928, it was found that treatments containing the furan ring occurred twice in the 4 highest in 1 experiment (Table 7), 4 times out of the 4 in another (Table 8), and 3 times out of the 4 in the other field experiment (Table 9).

PRELIMINARY OBSERVATIONS ON TWO SPECIES OF BEAUVERIA ATTACKING THE CORN BORER, PYRAUSTA NUBILALIS HÜBNER

C. L. LEFEBVRE[1]

INTRODUCTION

The purpose of this investigation was to note the morphological and physiological differences, if any, between certain species of Beauveria that attack the corn borer. One species of this fungus, occurring on the corn borer imported from Manchuria, killed from 80 to 90 per cent of the larvae, when these were subjected to conditions favorable for hastening the emergence of insect parasites for biological study. Large numbers of corn-borer larvae are imported annually from various countries by Government investigators interested in the control of this insect. By so doing they have established insect parasites in sufficient quantities to be of significance in the control of this pest in the United States. A somewhat similar species of this fungus has been reported on the chinch bug (2), the gipsy moth, and the satin moth, in the United States. The question therefore arises whether that species, first found parasitic on the corn borer in the European Corn Borer Laboratory at Arlington, Mass., is the same as the one already found on the chinch bug and other insects of this country. If so, why has it never heretofore been reported to occur on the corn borer, now so common in the United States? With the aim of casting light on this question the investigation was undertaken.

TAXONOMIC CONSIDERATIONS

The genus Beauveria in which these species belong is included in the tribe Verticillieae of the *Fungi Imperfecti*. It might be well at this time to state briefly some of the opinions and confused statements brought forward by different writers when referring to various representatives of this genus. For example, European writers have been using the generic names Botrytis Mich. and Sporotrichum Link when referring to members of this genus. Due to the confusion in naming members of these two groups, Vuillemin (10), 1912, instituted for them the new genus Beauveria; thus *Botrytis Bassiana* Bals. becomes *Beauveria Bassiana* (Bals.) Vuill. In

[1] The writer wishes to express his appreciation and gratitude to D. W. Jones, who has given him the opportunity to collaborate with Dr. Bartlett in this investigation.

He also wishes to acknowledge his indebtedness for helpful criticisms and suggestions to Professor William H. Weston, Jr., of Harvard University, under whom this work in its mycological aspects was carried on; and to Dr. W. H. Sawyer and Dr. M. T. Smulyan for cultures of *Beauveria globulifera* and *Isaria farinosa* (Dicks.) Fr., which they kindly furnished for comparative study.

1914 Picard (7) published a paper on entomogenous fungi in which he transferred Spegazzini's species *Sporotrichum globuliferum* to Vuillemin's genus, as *Beauveria globulifera*. From time to time others, chiefly Petch (5), Arnaud (1), and Billings and Glenn (2), have added considerable to our knowledge of these forms. This paper will be confined to a discussion of the two first-mentioned species.

The morphological differences between the species of Beauveria that are parasitic on insects are very slight, according to published reports by Petch and others. In the eight species of Beauveria described the main criteria used in separating them are the shape and size of spores, which are figured as being either oval or globose but may vary from these distinct extremes if the several species are grown on artificial media. Petch (5) makes clear this point in that, for *Beauveria Bassiana*, de Bary has described the conidia as being globular, 2.5 to 2.8 μ in diameter, while Delacroix records 2 to 2.5 μ as their dimensions, and Sawada has figured them as globose or broadly oval. Petch's own measurements, made from a culture secured from The National Collection of Type Cultures, records the conidia as being either broadly oval, 1.5 to 2.5 μ by 1.2 to 2 μ, or globose, 1.5 μ in diameter. Measurements of 250 conidia are summarized by the writer in table 4. In general, these measurements agree with those of Petch but, as they are tabulated in 0.5 μ classes and these, in turn, plotted on a curve of frequency distribution, the ranges in variation are more clearly brought out.

Since there is such a close similarity in spore size and shape between *Beauveria Bassiana* and *B. globulifera*, obviously other methods of separating these two organisms must be considered. Therefore, a more detailed study of the cultural characteristics on various artificial media and their virulence on the corn borer as a host was made by the writer, and the results are presented in this paper.

MATERIAL AND METHODS

The fungus, *Beauveria Bassiana*, was isolated from corn-borer larvae obtained from Dr. K. Bartlett, of the Corn Borer Laboratory, Arlington, Mass. For comparative study, cultures of *B. Bassiana* and *B. globulifera* were secured from the following sources: a culture of *B. Bassiana*, isolated by Dr. T. Petch, was obtained from the *Centralbureau voor Schimmelculture*, Baarn, Holland, and cultures of *B. globulifera* were obtained from Dr. W. H. Sawyer, Jr., and Dr. M. T. Smulyan, of the Gipsy Moth Laboratory, Melrose, Mass. Since these fungi grow very rapidly on potato-dextrose agar, it was relatively easy to secure and maintain a plentiful supply of inoculum.

In order to test the pathogenicity of *Beauveria Bassiana* and *B. globulifera* seven inoculation experiments were made during the course of the

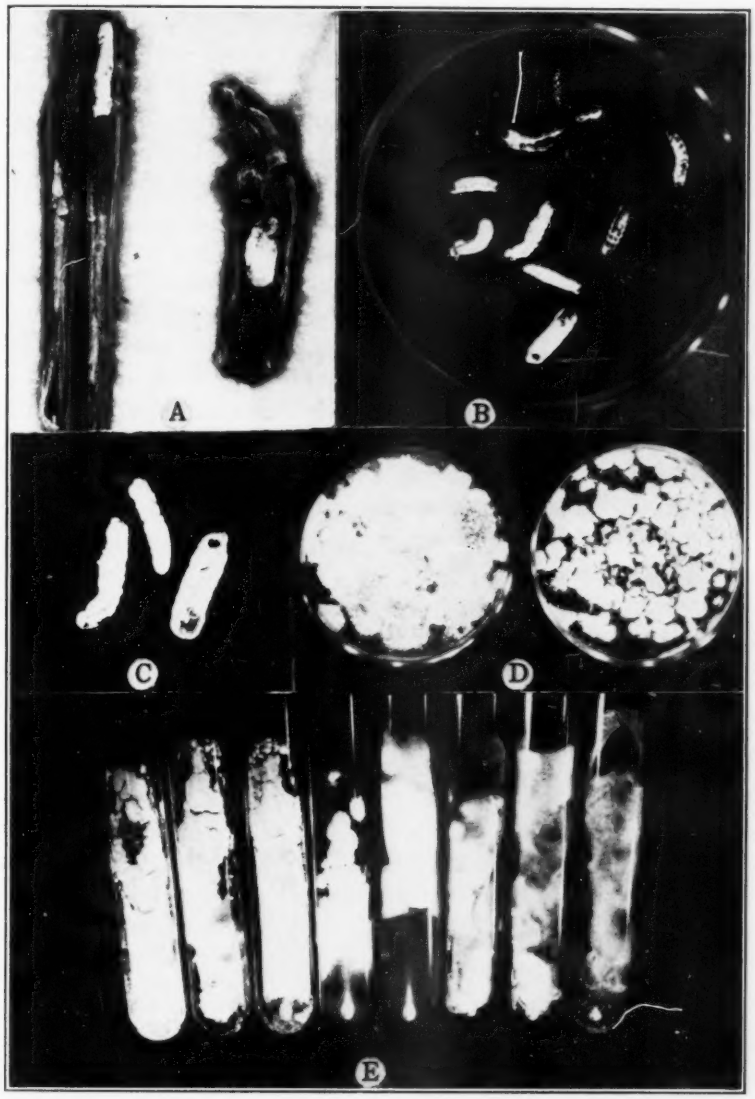

FIG. 1, A. Longitudinal sections of sweet-corn stems showing dead corn-borer larvae that were infected by dusting spores of *Beauveria Bassiana* on plants in the field. ×1. B. Corn-borer larvae killed within 2 days, after dusting with spores of *B. Bassiana*, in a moist chamber. ×⅔. C. Three dead corn borers showing larvae covered with white, mealy mycelium and spore mass of *B. Bassiana*. App. ×1. D. Left, *B. globulifera* on potato-dextrose agar showing elevated, cottony type of growth; right, *B. Bassiana* on potato-dextrose agar showing flat, mealy type of growth. ×½. E. Cultures of the two species on potato-dextrose agar, the left four showing the flat, mealy growth of *B. Bassiana;* the right four showing the elevated, floccose growth of *B. globulifera*. ×⅔.

summer, using as a host corn-borer larvae obtained through the courtesy of Dr. Bartlett. In each case a like number of larvae were used as controls.

Since, in the literature, there are accounts of the destructiveness of *Isaria farinosa* to various insects, it seemed desirable to test, along with these experiments, the possible pathogenicity of this fungus on the corn borer; therefore, comparative inoculation trials were made with this organism (Table 3).

Pure cultures of the fungus from infected corn borers are relatively easily secured after the larvae have been kept at room temperature for several days. During the course of this time a white mycelial outgrowth becomes evident, which turns later to a cream-colored, chalky, pulverulent mass of spores completely covering the larvae (Fig. 1, A and B). In this stage, by touching a sterile needle to this spore mass, a pure culture is readily isolated, or when isolating *Beauveria Bassiana* from corn-borer larvae collected in the field (Fig. 1, A) a minute bit of the fungus is easily scraped from the infected surface into tubes of melted potato agar, and these are poured into Petri plates, giving isolated colonies from which pure cultures may be made.

After the larvae have been dead for some time they become hard, brittle mummies; but, before any surface sporulation has occurred, isolations may be easily accomplished by dipping these in 95 per cent alcohol, quickly passing them through a flame, then cutting the larvae open with a sterile knife; the white mass of mycelium and spores can then be transferred in a relatively pure state to tubes of agar.

For comparative study *Beauveria Bassiana* and *B. globulifera* were grown in tubes of potato-dextrose agar and subjected to various temperatures. Similarly, these two organisms were grown in van Tieghem cells to determine the method and nature of spore germination. In all phases of this comparative study of the two organisms they were subjected to identical conditions.

GROWTH OF THE ORGANISMS ON ARTIFICIAL MEDIA

After observing the persistent, characteristic development of the stock cultures of these two organisms on potato-dextrose agar, it is obvious that *Beauveria Bassiana* always gives a mealy, chalky, pulverulent growth, while *B. globulifera* always produces an elevated, cottony, floccose growth (Fig. 1, D and E). It seemed advisable, therefore, to culture these fungi on various kinds of artificial media to see if these differences were lasting characteristics. The results are given in table 1.

From table 1 it is apparent that the fungus isolated from the corn borer always produces a characteristically flat, mealy, chalky, pulverulent growth on the various media used; while *Beauveria globulifera* produces a

TABLE 1.—*Summary of comparative development of Beauveria Bassiana and B. globulifera on artificial media. These two species were grown in the laboratory under identical conditions, and all transfers of each organism to these various substrata were made from the same stock culture*

Medium	Type of growth of B. Bassiana	Type of growth of B. globulifera
Potato-dextrose agar	Flat, chalky, mealy growth over whole slant	Loose, elevated, cottony growth over whole slant
[a]Bacto-prune agar	Flat, mealy, chalky growth. Slant ¾ covered	Loose, cottony, elevated to a less degree than above. Growth over whole slant
[a]Bacto-nutrient agar	Whole slant covered with flat, mealy, chalky growth. Agar purplish at base of slant	" " " "
[a]Proteose-peptone 2% agar added	Profuse, flat, mealy, pulverulent growth covering whole slant	Very profuse, elevated, loose, cottony growth over whole slant
[a]Bacto-peptone 2% agar added	" " " "	" " " "
[a]Lactose 2% agar added	Slight growth, slightly chalky. Lighter color than on proteose peptone	Slight growth, cottony, spheroid colony of hyphae
[a]Bacto-lactose 2% agar added	" " " "	Better growth than above, cottony, elevated
[a]Bacto-dextrose 2% agar added	" " " " and cottony	Slight growth, but elevated and cottony in patches
Corn meal	Little growth, lighter color than on proteose peptone	Little growth, slightly elevated, cottony colonies
[a]Bacto-bean-pod agar	Mealy, chalky growth. Base of slant well covered. Top covered in patches	Mealy, but somewhat elevated growth, of darker color than fungus from corn borer
[a]Bacto-malt agar	" " " "	Profuse, elevated, cottony growth, covering whole slant
Potato slab	Slightly cottony at first, soon becoming mealy, chalky, and flat	Profuse, elevated, cottony growth, in some cases turning potato slightly purple
Corn-meal mush	Flat, mealy, pulverulent growth	Profuse, elevated, cottony growth
Oatmeal	Slightly cottony at first, becoming flat, mealy, chalky	" " " "
10% gelatine	Flat, mealy growth. Gelatine turns brownish, amber color	Cottony, elevated growth

[a] Difco artificial media were used in this experiment.

characteristic elevated, cottony, loose, floccose growth, excluding the one culture on bean-pod agar, in which case the two organisms had a somewhat similar chalky appearance. Whether this character will change back again when the organisms are sown on an agar that normally gives an elevated, cottony growth has not yet been tried.

This consistently chalky, pulverulent growth on various media confirms the fact that this fungus, isolated from the corn borer imported from

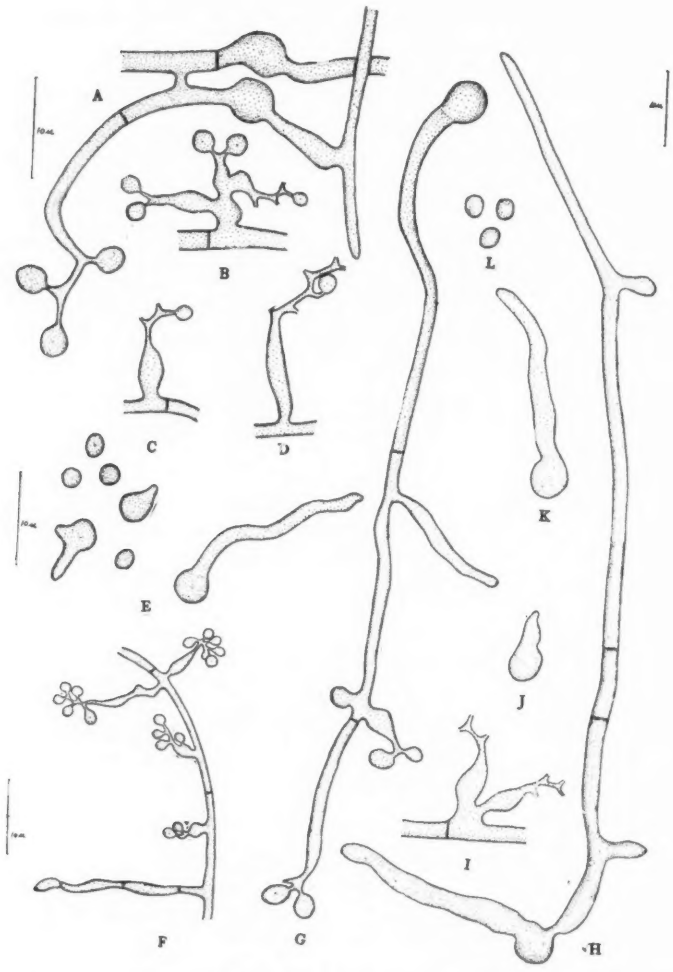

FIG. 2, A to G. *Beauveria Bassiana;* H to L. *B. globulifera.* A. Germinating spores with germ tubes anastomosing and one of them bearing conidia. ×1,350. B. Conidiophore branching into flask-shape phialides. ×1,250. C–D. Phialides: the one in C is flask-shape and that in D more slender, both having zigzag sterigmata. ×1,250. E. Four mature conidia, 3 showing pronounced swelling when germinating. ×1,250. F. Hypha bearing conidiophores and phialides; the spores are shown borne in clusters and singly. ×500. G. A germinating spore (after 32 hours) showing swelling of the conidia, and the septate, branching germ tube bearing conidia terminally and on a phialide. ×1,000. H. A germinating spore of *B. globulifera* (after 32 hours) showing swelling of the conidia, and the septate, branching germ tube; in this species, conidia are not yet formed. ×1,000. I. Conidiophore branching into flask-shape phialides showing zigzag sterigmata. ×1,250. J–K. Conidia showing pronounced swelling upon germination. ×1,250. L. Mature conidia at the time of shedding from the sterigmata. ×1,250.

Manchuria, is indeed *Beauveria Bassiana* (Bals.) Vuill. and is therefore distinct from *B. globulifera* (Speg.) Picard, common in this country. These findings agree very well with those of Petch and others who have done work on this group of imperfect fungi. This fungus, therefore, will be referred to as *B. Bassiana* throughout the remainder of this paper.

Another point of distinction between these two organisms, clearly brought out in the course of culturing them in the laboratory, was the difference in time required for spore formation. Observations on cultures of *Beauveria Bassiana* comprising data on 50 to 75 tubes of potato-dextrose agar grown under various conditions showed that abundant spore formation took place within a week, usually within 3 or 4 days; while in a like number of tubes of *B. globulifera* under identical conditions obvious spore formation did not take place until after the cultures had been growing 2 weeks to a month and then, only sparsely.

Spore germination: Studies of spore germination were made by placing conidia in drops of distilled water or 2 per cent peptone in van Tieghem cells following the method and percentage of germination. In 24 to 48 hours the spores swelled and put out one or more slender thin-wall germ tubes containing clear hyaline protoplasm. Frequently, they were found anastomosing (Fig. 2, A).

After 32 hours the germ tubes of *Beauveria globulifera* range from a few microns to as much as 200 μ in length, with the development of numerous branches 100 μ or more in length intertwining in a dense weft; while in *B. Bassiana* the germ tubes were much shorter, ranging from a few microns to about 80 μ, and the branches arising from these were very short. Consequently, the wefted appearance of the former was totally lacking in the latter. Thus, almost immediately in their early stages the mycelial development in van Tieghem cells substantiates what one finds when the two organisms are growing on agar, in that the tufted growth of *B. globulifera* and the more flattened growth of *B. Bassiana* are obvious.

At this stage of development conidial production is evident in *Beauveria Bassiana* (Fig. 2, G), while it is totally lacking in *B. globulifera* (Fig. 2, H). Later, when both organisms are producing conidia abundantly, they are borne in rather compact, globose heads, either on the main hyphal branches or on short laterals that are usually at right angles to the main axis, being much constricted at the base and terminating in an elongated, flask-shape conidiophore (phialide). Perhaps more commonly when these are at right angles to the main axis they occur in whorls and these also terminate in phialides and may bear branches in the same manner as those on the primary axis. Frequently this branching is repeated, forming compact heads whose make-up is difficult to follow (Fig. 3, A, B, C).

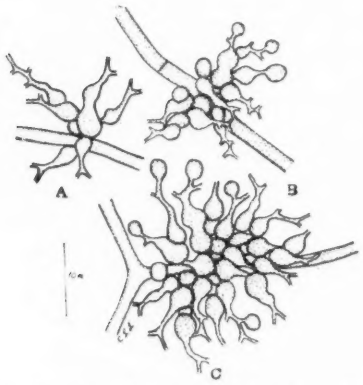

FIG. 3, A, B, and C. Hyphae bearing clusters of flask-shape phialides showing characteristic zigzag sterigmata. ×1,000.

Conidia are borne on short, slender, zigzag sterigmata borne at the apex of each phialide (Fig. 2, F). These, in turn, bear secondary sterigmata or short alternately arranged pedicels each of which bears a single conidium, and this may be repeated until a cyme of conidia is formed. These slender, zigzag, cymose sterigmata constitute the distinguishing character of the genus Beauveria (Fig. 2, C, D, and I).

In making observations on the percentage of spore germination, one was appalled at the small number of conidia that produced germ tubes, at all. The results of spore germination trials are given in table 2.

TABLE 2.—*Germination of spores of Beauveria Bassiana in distilled water and peptone*

Medium	Temperature	Per cent germination in center of droplet	Per cent germination edge of droplet
Steriie distilled water in 8 van Tieghem cells	Room	30, usually 10	90, unusual
Sterile distilled water in 6 Ward cells	Room	5	80 "
Sterile distilled water in 2 van Tieghem cells	10 C.	1 ±	1 ±
Sterile distilled water in 2 van Tieghem cells	27 C.	2	30
Sterile distilled water in 2 van Tieghem cells	33 C.	1 ±	1 ±
2 per cent peptone[a] in 2 van Tieghem cells	Room	50	90

[a] The writer has since secured much higher spore-germination percentages by placing the spores in peptone and dung decoctions.

Table 2 brings out the fact that the percentage of spore germination was somewhat higher when peptone was used as nutriment and that a greater number germinated around the edges of the droplet. When only a few spores were suspended in a droplet of the nutriment, germination seemed better; but too few observations were made to verify this fact, or it may be that the presence of large numbers of conidia in a droplet produces some antagonistic substances that have a tendency to lower the percentage of germination.

DIFFERENCES IN PATHOGENICITY OF BEAUVERIA BASSIANA, B. GLOBULIFERA, AND ISARIA FARINOSA

Because of manifest differences between *Beauveria Bassiana* and *B. globulifera* on artificial media and in the method of spore germination, it was thought that further contrasting characteristics might be obtained by trying inoculation experiments, using the corn-borer larvae as more or less of a differential host. Likewise, larvae were inoculated with *Isaria farinosa*, as this fungus is known to be parasitic on various insects (Table 3).

From table 3 it is apparent that there is a striking difference in the ability of these 3 organisms to infect the corn-borer larvae. Of the 50 larvae inoculated with *Beauveria Bassiana*, 30 died within 3 days when the larvae were thoroughly mixed and covered with spores by placing them in test tubes containing inoculum. By dusting, 6 out of 10 were killed within 5 days; and, when larvae were allowed to crawl over infected soil, 4 died within 6 days. In similar experiments in which 60 corn-borer larvae were inoculated with *B. globulifera* only 4 borers died from the disease. *Isaria farinosa* seemed to have no effect on the larvae. Controls in all cases showed no signs of infection. All these experiments were made at room temperature in the course of the summer, in the research laboratory of the Farlow Herbarium.

In another experiment 2 lots of 10 corn-borer larvae were dusted with spores of *Beauveria Bassiana* and *B. globulifera*, respectively; one lot was placed in the ice chest at 10–11° C. and the other placed in the 32–33° C. oven. There was no infection in either case, even though the 2 organisms grew on potato-dextrose agar at these temperatures. In all of these inoculation trials Petri dishes containing several layers of moist blotting paper were used as moist chambers, and string beans, thoroughly washed with sterile distilled water, were inserted in the chambers for food for the insects.

Arnaud (1) states that if silkworms, inoculated with *Beauveria Bassiana*, are kept in a moist chamber for 24 hours, then transferred to a dry container, the fungus will develop much better, and that if the infected

TABLE 3.—*Comparative pathogenicity of Beauveria Bassiana, B. globulifera, and Isaria farinosa on the corn borer in laboratory experiments*

	Date inoculated	Number inoculated	Method of inoculation	Date of infection noted	Number killed	Control	Remarks
Beauveria Bassiana	June 25	10	Dusting	June 30	6	No infection	4 others died but showed no disease
" "	July 6	10	Dusted surface of soil	July 12	4	"	
" "	" 12	10	Borers placed in test tube containing fungus	" 14	10	"	
" "	" 26	10	" " "	" 29	10	"	
" "	Aug. 1	10	" " "	Aug. 4	10	"	
" "	" 3	10	Dusting		0	"	Placed in ice chest −11° C.
" "	" 3	10	Dusting		0	"	Placed in 33° C. oven
B. globulifera	June 25	10	Dusting		0	"	
" "	July 6	10	Dusted surface of soil		0	"	
" "	" 12	10	Borers placed in test tube containing fungus		0	"	
" "	" 26	20	" " "		0	"	
" "	Aug. 1	10	" " "	Aug. 5	4	"	First time to be noted
" "	" 3	10	Dusting		0	"	Placed in ice chest −11° C.
" "	" 3	10	Dusting		0	"	Placed in 33° C. oven
Isaria farinosa	June 25	10	Dusting		0	"	
" "	July 12	10	Borers placed in test tube containing fungus		0	"	

worms are kept in a humid atmosphere continuously they will recover from the disease. This certainly does not hold true in the case of the corn borers, for, once they become infected, as first manifested by their becoming rather sluggish, failing to respond readily to external stimuli, and in some instances turning a "pinkish" color, none of these has ever been seen to recover.

Although only a few field experiments have been made, they indicate that *Beauveria Bassiana* is an organism possessing favorable potentialities for at least partial control of the corn borer in that the fungus readily infects and is destructive to the larvae yet grows well on artificial media and produces spores in vast numbers. Moreover, the corn-borer larvae, ensconced within the succulent tissues of the host during most of their period of feeding, live under relatively moist conditions, ideal for infection with fungous parasites. They are thus advantageously in contrast to certain other destructive insects against which fungous diseases have been used with little success on account of their feeding in the open under drier conditions. A summary of these field experiments and subsequent ones will shortly be compiled by Dr. K. Bartlett.

Spore characters: Since in the literature cited there is such a lack of agreement as to the size and shape of spores of *Beauveria Bassiana* and *B. globulifera*, measurements of 250 conidia were made by the writer, and the results were given in the following table.

TABLE 4.—*Summarized comparative measurements of conidia of Beauveria Bassiana and B. globulifera in water*

Length in μ	Classes *B. Bassiana*	*B. globulifera*	Diameter in μ	Classes *B. Bassiana*	*B. globulifera*
1.0–1.5	2	1	1.0–1.5	8	6
1.6–2.0	40	68	1.6–2.0	84	87
2.1–2.5	121	113	2.1–2.5	120	122
2.6–3.0	69	50	2.6–3.0	36	30
3.1–3.5	12	13	3.1–3.5	2	4
3.6–4.0	5	1	3.6–4.0	0	1
4.1–4.5	1	1			
4.6–5.0	0	1			
5.1–5.5	0	1			
5.6–6.0	0	1			

One notes at once that the larger number of spores fall in the class 2.1–2.5 μ for both length and diameter measurements. In general, the spores are globose but vary somewhat, as shown. Other writers give the following measurements:

Conidia globose:

Beauveria Bassiana (Bals.) Vuill.—

Conidia 2.5–2.8 μ (de Bary) (3)
" 2.0–2.5 μ (Delacr.) (4)
" 1.5–2.5 × 1.2–2.0 or globose
" 1.5 μ diam. (Petch) (5)

Beauveria globulifera (Speg.) Picard—

Conidia 2.0–2.5 × 1.5–2.0 μ (Speg.) (8)
" 1.74–2.5 μ (Pettit) (6)

From these data it is quite evident that spore size and shape are not a sound basis on which to distinguish *Beauveria Bassiana* from *B. globulifera* (Fig. 4).

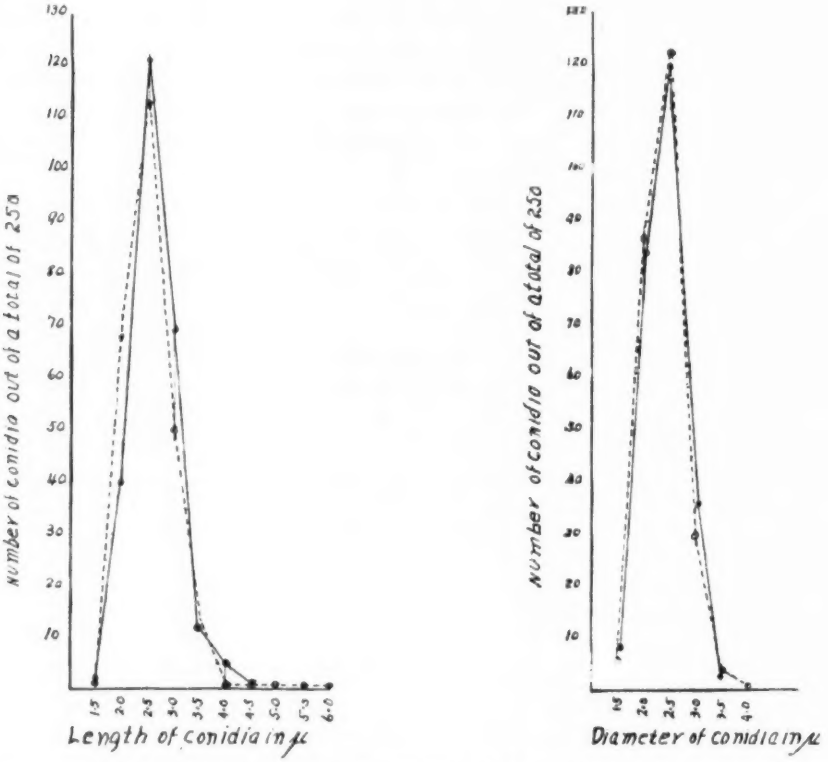
Number of conidia out of a total of 250
Length of conidia in μ
Number of conidia out of a total of 250
Diameter of conidia in μ

FIG. 4. Graphs showing comparative dimensions of the conidia of *Beauveria Bassiana* (———) and *B. globulifera* (– – – –) when grown and measured under identical conditions.

PRESENT KNOWN DISTRIBUTION OF THE FUNGUS

Beauveria Bassiana is known to be fairly wide-spread, as it occurs on insects in many localities, such as Europe, China, Japan, Ceylon, the Phil-

ippine Islands, and Manchuria. In fact, the fungus is so generally prevalent that it has been found impractical to ship corn-borer larvae from the district of Kanto, Manchuria, to the United States, where they are used to study the emergence of insect parasites. As far as the writer is aware, *B. Bassiana* has never been known to occur on the corn borer in the United States. The only record of this fungus in the United States is by Van Hook (9), who states that he found *Botrytis Bassiana* on a dead insect fast to a living leaf of cultivated strawberry; but, apparently, there is no mention that he cultured the fungus, nor is material available whereby further check on this determination can be made.

PERSISTENCE OF THE FUNGUS

The writer has not made extensive observations on the retention of vitality of the fungus, noting only that cultures kept in the laboratory 10 months are still capable of infecting the corn-borer larvae. This is in keeping with Arnaud (1), who states that by making frequent transfers cultures of *Beauveria Bassiana* were kept in the laboratory 2 years without altering their ability to infect the silkworm.

SUMMARY

Comparable cultural and inoculation experiments made with *Beauveria Bassiana* and *B. globulifera* have cast light of certain significance on the identity of these fungi (1) when grown on artificial media and (2) when used to inoculate corn-borer larvae.

Pure cultures of *B. Bassiana* are relatively easily secured, due to its fairly rapid growth and abundant spore formation.

On artificial media *B. Bassiana* is characterized by producing a flat, mealy, chalky, pulverulent growth, while *B. globulifera* forms an elevated, cottony, floccose growth.

In culture and on the corn borer *B. Bassiana* produces conidia much sooner and in greater numbers than *B. globulifera*.

On various agars and in van Tieghem cells *B. globulifera* has a greater mycelial development.

Beauveria Bassiana is a much more virulent pathogen on the corn borer than is *B. globulifera*.

For comparison with the two species of Beauveria, *Isaria farinosa* was tested under similar conditions but proved to be noninfectious.

Quantitative measurements of 250 conidia are given in tabular form, bringing out the close similarity in size and shape of the conidia of *B. Bassiana* and *B. globulifera*.

As far as is known, *B. Bassiana* has never been reported to occur on the corn borer in the United States but has been collected once in this country.

In cultures *B. Bassiana* does not seem to lose its virulence readily, at least not within the course of 10 months.

LABORATORIES OF CRYPTOGAMIC BOTANY,
HARVARD UNIVERSITY,
CAMBRIDGE, MASS.

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BLIGHT OF CARROTS CAUSED BY SCLEROTIUM ROLFSII, WITH GEOGRAPHIC DISTRIBUTION AND HOST RANGE OF THE FUNGUS

GEORGE F. WEBER

A blight of carrots caused by *Sclerotium rolfsii* Sacc. was found near Gainesville, Fla., during the early summer of 1928. The disease appeared to be spreading in both directions in the rows from points of primary infection, killing the plant successively. This disease of carrots has not been previously described, so far as the writer has determined, although numerous hosts of the fungus have been reported.

DESCRIPTION OF THE DISEASE

The earliest symptoms of infected carrots are the general yellowness and lack of turgidity of the old leaves. As the disease develops these symptoms spread to the younger leaves, until, finally, all of them become involved. This development may require only 2 or 3 days under favorable conditions, yet the period is long enough to show distinctively the various stages in the advance of the disease. The older leaves wilt completely and become prostrate (Figs. 1 and 2), followed, successively, by the younger ones, until all of them lie on the soil surface. In this stage the leaves are loosely attached to the crown, and the root cannot be removed from the soil by pulling the leaves, while healthy roots are easily removed in this

FIG. 1. Portion of a carrot field showing more than 50 per cent of the plants affected with *Sclerotium rolfsii* Sacc.

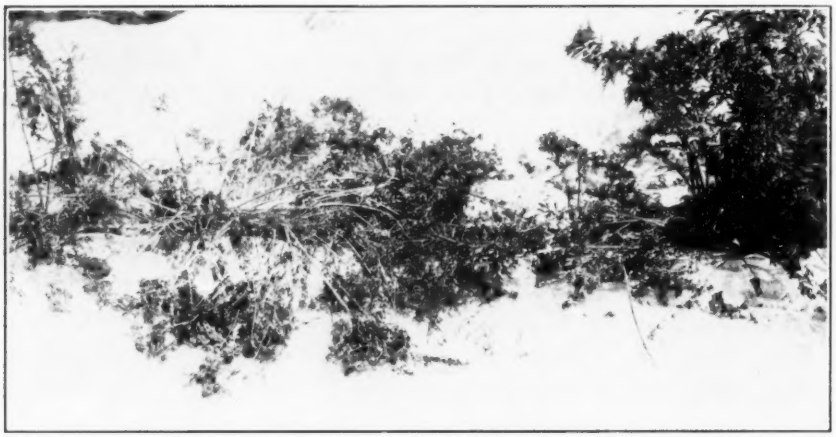

FIG. 2. Section of carrot row showing diseased plants compared with healthy ones at reader's right.

manner. They then lose their green or yellow color and become brown and crisp. The petioles easily become loosened from the crown of the plant and may be scattered by the wind, leaving no trace of the plant or the disease. There are no other symptoms above the soil surface by which the disease is characterized, except in fields where the plants have not been properly cultivated and portions of the crown are exposed. If such exposed roots are attacked, they often show white mycelial growth of the parasite. In contrast with this characteristic of the disease on carrots, the writer has observed that on a large number of other host plants that are vigorously attacked the causal fungus usually overgrows the stems of the plant from the soil line to several inches above. This condition has been reported by Higgins (15), McClintock (17), and Miller (19). Takahasi (36) also noted the mycelium of a similar fungus advance 6 inches up larkspur stems. Wolf (43), however, reported that the mycelium was entirely underground on peanuts, in Alabama.

The fungus invades the entire root of the carrot very quickly, causing a wet rot (Fig. 3). In some instances, where the decay had advanced through the periderm, cortex, phloem, and cambium, the central cylinder of pits and woody tissue would easily slip out when the leaves were pulled, leaving the outer cylinder of cortex and phloem in the soil. This outer portion of the carrot was held in the soil by the mycelium of the fungus, visible in the soil to a distance of an inch or more around the carrot. The distance the mycelial growth penetrated the soil was apparently determined by the amount of moisture supplied to the soil by the decaying carrot. The soil was usually wet from 1 to 2 inches around the carrots, which were from 1

FIG. 3. The same plants shown in figure 2 removed from the ground and showing wet, soft decay.

to 2 inches in diameter. The distance around the smaller ones was relatively less. This condition appears to be of great importance in the spread of the disease in the row, as it enables the fungus to pass readily from one plant to another, even though the roots are not in direct contact. In advanced stages the decayed carrots dry out and shrink, leaving a cavity in the soil. The walls of the "sand-wells" thus found are held quite securely by the dried mycelium of the fungus. The sclerotia of the fungus are produced in large numbers on the deteriorating carrots in these cavities. Sclerotia were observed 6 inches below the soil surface, although not underground. Taubenhaus (37) recommended plowing 5 inches deep for the control of *Sclerotium rolfsii*. Under the above circumstances, however, such methods would probably place sclerotia on the surface rather than deep under the soil.

INOCULATION EXPERIMENTS

The fungus was obtained in pure culture by (a) planting mature sclerotia, which had been sterilized, on poured agar plates; (b) by transferring bits of the fluffy white mycelium that developed on infected roots (Fig. 4), kept for 24 hours in a moist chamber; and (c) by planting bits of invaded host tissue that had been surface sterilized. Fungus growth was evident on the plates after 12 hours. Within 4 days sclerotia began to develop. The mycelium and sclerotia (Fig. 5), in growth habits, color, size, and shape, were similar to descriptions given to *Sclerotium rolfsii* by Earle (9), Higgins (15), Rolfs (28, 29, 30), Saccardo (33), and Taubenhaus (37). Two weeks after the plantings were made in Petri dishes, the sclerotia produced therein were collected and used to inoculate carrots. Inocula-

FIG. 4. A naturally infected carrot showing point of attack and the development of the fungus during a 24-hour period in a moist chamber.

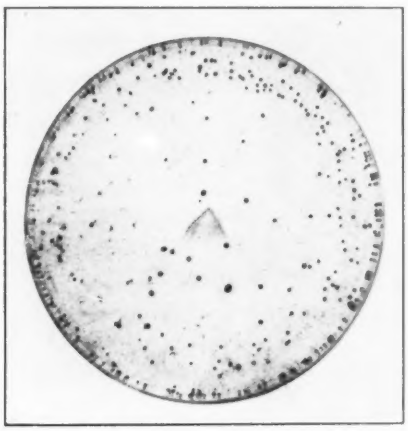

FIG. 5. *Sclerotium rolfsii* Sacc. in pure culture showing sclerotia.

tions were made in the field and in the laboratory, both with and without injury. A disease produced on inoculated plants was similar to that observed in the field.

TABLE 1.—*Results obtained by inoculating carrots with Sclerotium rolfsii from sources indicated*

Sources of inoculum	Parts inoculated	Number inoculated	Number inoculated	Checks	
				No.	Diseased
Sclerotia from field	Root	6	6	2	0
	Growing plant	12	10	2	0
Sclerotia from culture	Root	6	6	2	0
	Growing plant	12	11	2	0
Mycelium	Root	6	6	2	0
	Growing plant	12	6	2	0

In all cases of infection sclerotia were produced that were indistinguishable from those of the original culture. In moist chambers the inoculated carrots became overgrown with mycelium very rapidly. If they were left uncovered the mycelium developed more slowly but was much more dense. (Fig. 6).

FIG. 6. A naturally infected carrot (above) producing the disease on the healthy carrot (below) by contact, in an open dish.

The disease has been collected in Florida on a large number of hosts, as listed hereafter, and in most instances pure cultures were made. A series of cross inoculations was made in which the fungus from various hosts was used to inoculate healthy plants, once reported as hosts. Table 2 shows the source of the inoculum used in the experiment, although the sclerotia employed as inoculum were developed in artificial culture. The results are given in the form of fractions in which the denominator represents the number of plants or plant parts inoculated and numerator the number of

TABLE 2.—*Results obtained by inoculating plants with Sclerotium rolfsii obtained from sources indicated*

Source of fungus	Plants inoculated												
	Bean	Beet	Carrot	Cotton	Cucumber	Eggplant	Lettuce	Pea	Pepper	Potato	Squash	Tomato	Watermelon
Bean	8[a]/10	4/6	5/10	4/10	7/10	4/6	6/6	4/4	4/6	4/4	6/10	10/10	4/6
Beet	4/6	8/10	6/6	4/4	7/8	6/10	4/4	6/6	5/6	2/4	4/6	6/6	5/6
Carrot	5/6	7/10	20/20	4/6	6/6	4/6	6/6	4/6	6/6	4/4	6/6	6/6	5/6
Cotton	3/6	8/10	10/10	10/20	5/6	5/6	6/6	5/6	6/6	3/4	5/6	5/6	6/6
Cucumber	3/6	4/6	8/10	4/4	6/10	4/4	6/6	4/4	4/5	3/4	4/4	4/5	4/4
Eggplant	4/6	4/4	6/6	4/6	4/10	10/10	6/6	6/6	—	6/6	5/5	6/6	2/6
Lettuce	6/6	3/4	3/4	4/5	1/4	2/5	10/10	4/6	—	3/4	3/7	4/4	5/5
Pea	4/6	4/6	8/10	4/4	4/4	4/6	10/10	10/15	—	2/2	3/4	7/7	4/6
Pepper	6/6	4/4	10/10	4/4	4/6	4/5	4/5	1/4	10/10	4/6	6/6	6/10	6/10
Potato	5/6	4/4	6/6	3/6	3/4	6/6	5/5	6/7	—	10/10	4/4	4/6	3/5
Squash	6/6	4/6	8/8	6/6	5/6	5/6	3/4	4/5	3/6	3/4	15/15	7/8	4/6
Tomato	6/6	3/5	10/10	4/4	4/6	6/6	6/6	3/3	6/8	4/4	6/10	15/15	5/10
Watermelon	4/6	1/4	6/10	4/4	3/3	5/5	7/8	4/6	10/10	3/6	4/4	4/4	10/10

[a] The denominators of the fractions designate the number of inoculations and the numerators the number of infections.

plants that became infected. The number of plants inoculated may include seedlings, grown plants, or the fruits or roots of plants. Check plants, not shown in the table, were used throughout the experiment. None of the plants was intentionally wounded in the inoculation work, but it is possible that microscopic wounds were present on such hosts as potato and carrot. In most cases sclerotia constituted the inoculum, although mycelium was used occasionally. The inoculated plants were grown in the greenhouse and in field plots.

These results show that the fungus is exceedingly cosmopolitan. The strain isolated from each of 13 plants was capable of infecting the other 12. There was no consistent difference among the strains, even though there was some variation in size, shape, and color of the sclerotia both in culture and on the various hosts, and, consequently, they are considered the same species.

GEOGRAPHIC DISTRIBUTION

A blight of various plants caused by *Sclerotium rolfsii* has been reported in certain foreign countries. In the Western Hemisphere the fungus and the disease it causes have been reported from the following: Porto Rico (16, p. 210), (35), Cuba (31), Hawaii (7), and southern portions of the United States (9, 15, 28, 37, 39). There is little doubt of its occurrence in the other subtropical and tropical lands of North and South America that are subject to excessive rainfall under high summer temperatures. Available reports of this fungus and its damage to living plants show that it occurs in Egypt (5), South Africa (20), India (8), (18), Australia (4), Japan (21), Sumatra (23), Philippines (2), Java (11), and Ceylon (3). In fact, the general group of lands thus reporting the disease are apparently those in the tropical and subtropical countries where high temperatures prevail during the rainy season. This belt, with slight variations both north and south, roughly follows the Equator, around the globe. The disease undoubtedly occurs and causes considerable damage in other countries located in this tropical or subtropical belt that are subject simultaneously to abundant rainfall and high temperatures. Definite reports from all such lands, however, are not available.

In the United States the disease is general in the Southern States from Texas to the Carolinas and north to the Ohio River, with occasional reports from scattered Central States. In Florida this disease is common and widespread on a large number of hosts covering the entire State, on its various types of soil. Blight is generally worse on the light, sandy types of soil during the rainy periods of summer, but this is not always true, since pepper and tomato fields have suffered considerable damage during January and February, on the heavy muck, marl, and loam soils.

HOST RANGE

Since the reports of the disease on different host plants are widely scattered in the literature it seems worth while to compile a list of the hosts reported from the United States and foreign countries.

The fungus has been collected on a large number of hosts. These hosts are here reported alphabetically, listing the Florida collections as well as others reported in the literature. The new Florida hosts, herewith reported for the first time, lack reference numbers.

HOSTS

Scientific name	Literature reference	Scientific name	Literature reference
Allium cepa L.	1	*Chrysanthemum* sp.	29
" *sativum* L.	1	*Cichorium endivia* L.	22
Alocasia cucullata Schott	3	*Cinchona* sp.	27
Amaranthus retroflexus L.	29	*Citrullus vulgaris* Schrad.	29
" *spinosus* L.	29	*Citrus aurantifolia* (Christm.) Sur.	26
Ambrosia artemisiifolia L.	29	" *decumana* L.	—
Amygdalus persica L.	1	" *grandis* Osbeck	—
Anacardium occidentale L.	—	" *limonia* Osbeck	26
Antirrhinum majus L.	—	" *paradisi* MacF.	26
Aquilegia sp.	40	" *sinensis* P.	26
Arachis hypogaea L.	29	*Cochranea anchusaefolia* (Poir) Gürke	—
Areca catechu L.	6	*Coffea* sp.	26
Arrhenatherum elatius (L.) Beauv.	39	*Coleus blumei* Benth.	22
Artocarpus communis Forst	35	*Colocasia esculenta* Schott	—
Asimina sp.	25	*Commelina* sp.	22
Avena sativa L.	40	*Coreopsis lanceolata* L.	—
Bambusa vulgaris Schrad.	35	*Cosmos* sp.	40
Bauhinia alba Buch-Ham	6	*Crataegus* sp.	—
Begonia sp.	22	*Crotalaria anagyroides* H. B. & K.	11
Beta vulgaris L.	29	" *juncea* L.	11
" *vulgaris cicla* Moq.	40	" *sericea* Retz	—
" *vulgaris macrorhiza* Stev.	1	" *spectabilis* Roth	—
Brassica juncea (L.) Cosson	25	" *usaramoensis* E. G. B.	—
" *oleracea* L.	29	*Cucumis melo* L.	—
" *oleracea acephala* (DC.) Kales	40	" *sativis* L.	—
" *oleracea botrytis* L.	—	*Cucurbita pepo condensa* Bailey	29
" *rapa* L.	—	*Cyamopsis tetragonoloba* Taub.	40
Bryophyllum pinnatum Kurz.	22	*Cynara scolymus* L.	5
Caladium bicolor Vent.	—	*Cynoglossum* sp.	—
Calendula officinalis L.	40	*Dahlia pinnata* Cav.	—
Callistephus chinensis Nees	—	*Daphne odora* Thunbg.	29
Campanula carpatica Jacq.	24	*Datura metel* L.	—
" *medium* L.	24	*Daucus carota* L.	—
" *nobilis* Lindl.	24	*Delphinium ajacis* L.	—
" *persicifolia* L.	24	" *grandiflorum* L.	—
Canavalia ensiformis DC.	14	*Desmodium molle* DC.	29
" *gladiata* DC.	23	" *tortuosum* DC.	29
Cannabis sativa L.	1	*Dianthus barbatus* L.	—
Capriola dactylon Kze.	—	" *caryophyllus* L.	—
Capsicum annuum L.	29		
Chenopodium sp.	22		

HOSTS

Scientific name	Literature reference
" *plumarius* L.	24
Dioscorea sativa L.	4
Dolichos multiflorus T. & G.	22
Dracocephalum argunense Tisch.	24
Duranta plumieri Jacq.	—
Echinochloa frumentacea Link	22
Eleusine coracana Gaertn.	18
Erechtites hieracifolia (L.) Raf.	29
Erigeron canadensis L.	29
" *glabellus* Nutt.	24
Eupatorium ageratoides L.	24
Ficus carica L.	29
" *elastica* Roxb.	41
Forsythia sp.	40
Fragaria americana Brit.	—
" *virginiana* Duche	—
Gladiolus sp.	—
Glycine hispida Max	2
Gossypium hirsutum L.	—
Helianthus annuus L.	1
" *tuberosus* L.	—
Hibiscus cannabinus L.	13
" *esculentus* L.	25
Hippeastrum equestre Herb.	—
Holcus sorghum L.	1
Hordeum sativum Jess.	22
Hosta lancifolia Tratt.	40
Hydrangea paniculata Sieb.	29
Impatiens sultani Hook.	22
Indigofera endecaphylla Jacq.	34
" *hirsuta* Jacq.	34
Ipomoea batatas (L.) Lam.	29
" *purpurea* (L.) Roth	29
Iris sp.	40
" *xiphium* L.	—
Jasminum floridum Brenge	—
Lactuca sativa L.	—
Lagenaria leucantha Rusby	2
Lathyrus odoratus L.	—
Lespedeza simulata M. & B.	—
Lippia canescens Kunth	11
Lycopersicum esculentum Mill.	29
Malus sylvestris Mill.	4
Medicago sativa L.	1
Melilotus alba Desr.	22
Mimosa invisa Mart.	34
Musa sapientum L.	4
Myriophyllum sp.	22
Myrtus communis L.	—
Narcissus tazetta L.	—
" *tazetta orientalis* Hort.	—
Nicotiana rustica L.	20
" *tabacum* L.	42
Ornithogalum umbellatum L.	22
Oryza sativa L.	32
Oxalis sp.	22
Passiflora incarnata L.	—
Pentstemon murrayanus Hook.	24
" *pubescens* Soland.	24
Persea americana Mill.	—
Petroselinum hortense Hoffm.	22
Phaseolus vulgaris L.	29
Phlox subulata L.	24
Physalis angulata L.	—
" *pubescens* L.	—
Piper betle L.	38
" *nigrum* L.	—
Pisum sativum L.	29
Pittosporum tobira Ait.	—
Pteris longifolia L.	22
Raphanus sativus L.	3
Raphiolepis indica Lindl.	—
Reseda odorata L.	22
Rheum rhaponticum L.	29
Richardia scabra St. Hil.	—
Ricinus communis L.	22
Rosa rugosa Thunbg.	—
Rudbeckia laciniata L.	—
Saccharum officinarum L.	—
Salvia officinalis L.	22
Scabiosa atropurpurea L.	—
Scrophularia sp.	—
Sida rhombifolia L.	—
Soja max Piper	40
Solanum aculeatissimum Jacq.	—
" *gracile* Link	—
" *melongena* L.	29
" *mexicanum* D. & P.	—
" *munistrum* BiH.	—
" *nigrum* L.	—
" *tuberosum* L.	29
Stizolobium deeringianum Bort.	—
Tagetes sp.	—
Thymus vulgaris L.	22
Trifolium incarnatum L.	1
Triticum aestivum L.	10
Tropaeolum sp.	22
Vicia faba L.	35
Vigna catjang Walp.	1
" *hoei* Jacq.	34
" *oligosperma* Jacq.	—
" *sinensis* (L.) Endl.	3
Viola odorata L.	29
" *tricolor* L.	—
Xanthosoma sagittifolium (L.) Schott	12
Zantedeschia aethiopica Spreng.	—
Zea everta Sturt.	22
" *mays* L.	—
Zinnia elegans Jacq.	—

This list includes only a single report of each host. Where the host has been found in Florida, no other records are given. These hosts include succulent and woody, annual and perennial, and monocots and dicots. The fungus is more common on the succulent annuals, and it is not usually found on the woody perennials except under the most favorable temperature and moisture environment.

SUMMARY

1. A description of the disease caused by *Sclerotium rolfsii* Sacc. attacking carrots is given.
2. Isolations and inoculation experiments were conducted proving its pathogenicity.
3. Cross inoculations were conducted with 13 strains of the fungus, isolated from as many different plants, all of which proved susceptible to the various strains.
4. The strains of the fungus, although showing some variation in culture and on the hosts, did not warrant recognition.
5. The geographical distribution and host range of the fungus are given. One hundred and eighty-nine host plants are listed, 68 of which are reported here for the first time.

FLORIDA AGRICULTURAL EXPERIMENT STATION,
GAINESVILLE, FLA.

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NEW APPLE-ROT FUNGI FROM WASHINGTON

GEORGE D. RUEHLE[2, 3]

Beginning in the fall of 1926 and continuing through 1927, 1928, and 1929, a detailed study was made of the fungi causing decay of Washington apples in cold storage. Apples from representative districts obtained from commercially packed lots and placed in cold storage were given systematic examinations at intervals during their storage life and isolations of fungi made from the lesions which appeared. A total of 1,118 isolations have been studied. The following fungi have been isolated and proved by inoculation tests to be capable of causing decay either at cold-storage or higher temperatures:

PHYCOMYCETES

Mucor piriformis Fischer
Rhizopus nigricans Ehr.

ASCOMYCETES

Pleospora fructicola (Newton) Ruehle
Mycosphaerella tulasnei Janez.

FUNGI IMPERFECTI

Phoma, No. 1
" No. 2
Coniothyrium, No. 1
" No. 2
Microdiplodia, sp. undet.
Gloeosporium perennans Z. & C.
Pestalozzia hartigii Tub.
Coryneum foliicolum Fckl.
Oospora, sp. undet.
Cephalosporium carpogenum, n. sp.
Penicillium expansum Lk.
" *puberulum* Banier
" *verrucosum* Biourge
" *olivino-viride* Biourge

[1] Published with the approval of the Director of the Washington Agricultural Experiment Station as Scientific Paper No. 187, College of Agriculture and Experiment Station, Pullman, Washington.

[2] Formerly Research Assistant in Plant Pathology, Washington Agricultural Experiment Station, Pullman, Washington.

[3] The writer wishes to express his deep appreciation to Dr. F. D. Heald for able counsel and constructive criticism during the course of the work and for help in preparation of the manuscript.

Penicillium viridicatum Westling
" *martensii* Biourge, and 5 other unidentified species
Sporotrichum malorum Kidd & Beaum.
Sporotrichum carpogenum, n. sp.
Botrytis cinerea Pers.
Botrytis mali, n. sp.
Cladosporium malorum, n. sp.
" *herbarum* Lk. (See *Mycosphaerella tulasnei*)
Hormodendron cladosporioides (Fr.) Sacc.
Stemphylium congestum Newton
" " Newton, var. *minor* Ruehle
" (See *Pleospora fructicola*)
Alternaria tenuis Nees.
" *mali* Roberts
" No. 3
" No. 4
" No. 5
Fusarium No. 1
" No. 2
Ramularia magnusiana (Sacc.) Lind.
" No. 2
Epicoccum granulatum Penz.

BASIDIOMYCETES

Corticium centrifugum (Lev.) Bres.

It is not the purpose of this article to present the details of this study but to describe certain new species, to record some new data on previously described species, and to give brief attention to some not previously recorded as causing decay of apples.

NEW SPECIES

Cephalosporium carpogenum, n. sp. A species belonging undoubtedly in the genus Cephalosporium was found to be one of the less frequent weak parasites of apple fruit. It was obtained from small, shallow, dark brown, firm areas bordering worm holes or punctures. When reinoculated into ripe Jonathan apples held at about 20° C., small, shallow, firm spots were formed in 30 days. At the end of 2 months, a few of these spots had increased to 20 mm. in diameter on the surface, but most of them remained smaller, although the apples were very ripe. Pathogenicity was not determined at cold-storage temperatures.

The fungus develops slowly on culture media. On 2 per cent dextrose-potato agar, the colonies are white to faintly pink at first and become light gray in age (Fig. 1, A). The development of the pink color seems to be

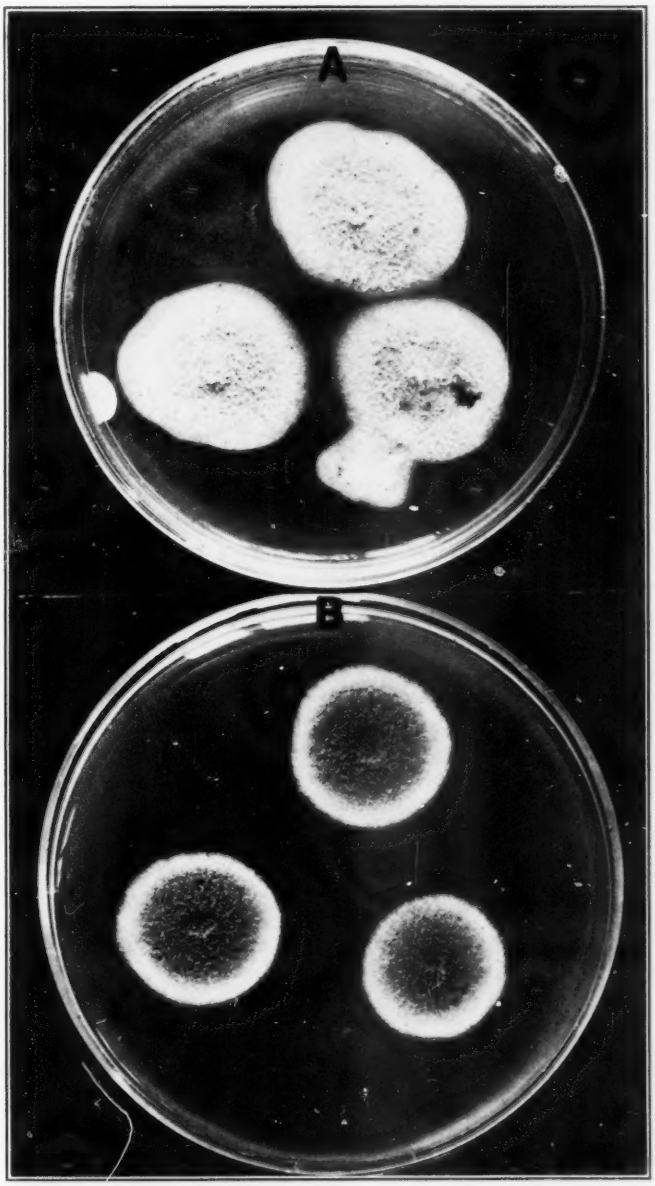

FIG. 1. A, *Cephalosporium carpogenum* on 2 per cent dextrose-potato agar; B, *Cladosporium malorum* on 2 per cent dextrose potato agar.

favored by growing the colonies in the light. The surface appears coarsely flocculate, bordered by a narrow zone of prostrate radiating hyphae. The hyphae are hyaline, sparsely branched, delicate, 1 to 2 μ in diameter, with septations very indistinct. In the aerial mycelium they are united into coarse strands composed of many parallel, adherent hyphae. The conidiophores are hyaline, simple, nonseptate, 1.5 to 2 μ wide at the base, and gradually attenuated toward the tip. They are 25 to 45 μ long and arise at nearly right angles from the hyphae. The conidia are hyaline, continuous, ellipsoidal to short cylindric, 4 to 8.5 μ long by 1.4 to 2.8 μ wide, produced successively on the apex of the conidiophores, but collecting there to form small, globose heads 8 to 15 μ in diameter, which break up readily in a water mount.

A single species of Cephalosporium has been previously reported from apple fruit in England by Kidd and Beaumont (6). This species, which they named *C. malorum* K. and Beaum., was always isolated associated with other fungi and there is nothing in their report to indicate that inoculation experiments were carried out with the fungus. In addition, the same investigators report two species of Hyalopus from apple fruit. Hyalopus is distinguished from Cephalosporium mainly by the tendency of the spore head of the former to remain intact, due to the greater amount of mucus formed. It has been suggested by Lindau (7) and Buchanan (1) that the two genera be combined, because this character is not considered sufficiently striking to warrant a separation.

A culture of *Cephalosporium malorum* was obtained from the Centraalbureau voor Schimmelcultures and was found on comparison to be distinct from the Washington fungus. The colonies develop much slower on culture media, and the spores are smaller (4 by 2 μ) in the English species. The Washington species also differs in spore characters from the two Hyalopus forms reported on the apple. It appears to be very similar to *C. charticola* Lindau, which has been described as occurring on moist paper, but the description of the latter is so inadequate that it is not possible to establish the identity of the two. It is, therefore, deemed advisable to consider the Washington fungus a new species, for which the name *Cephalosporium carpogenum* is proposed.

Sporotrichum carpogenum, n. sp. This form was isolated from a small, dark brown, firm, lesion on a Jonathan apple. It differs more or less distinctly from *S. malorum* (2), both in cultural characters and in size and shape of spores.

The colonies on 2 per cent dextrose-potato agar at about 20° C. spread very slowly, attaining a diameter of approximately 20 mm. in 10 days. In young colonies there is more or less fluffy overgrowth in the center, but later the aerial hyphae spread uniformly, forming a dense velvety mat of

a deep grayish olive (Ridgway XLVI) which becomes radiately furrowed in age. The margins are erose and without the border of white prostrate hyphae characteristic for *Sporotrichum malorum* (Fig. 2, C). The reverse side is dark olive buff to greenish olive in the deeper portions.

The hyphae are hyaline to light brown, 1 to 4 μ in diameter, with branches arising at short intervals. The hyphae are partly united into brown, coarse, strands of 3 to many parallel adherent hyphae. No loops or coils were observed in the aerial hyphae.

The conidiophores arise either singly as lateral outgrowths, usually at nearly right angles from the main hyphae, or more frequently in clusters of 2 to many arising at acute angles from short lateral branches from the main hyphae. Typically they are from 6 to 10 μ long and are swollen at the center and again at the tip to resemble a tenpin in shape, but occasionally the single conidiophores are much longer (up to 30 μ) and are scarcely distinguishable from the hypha from which they arise.

The conidia are subglobose to elliptical, hyaline, continuous, 2.8 to 6.4 by 1.8 to 3.5 μ. They are cut off successively from the apex of the conidiophore, and, in the aerial mycelium, accumulate about the tip of the conidiophore in a loose clump or "head," which breaks up readily in a water mount.

Because of the difference in rate of growth, character of colony, and size and shape of spores, this form is considered to be distinct from *Sporotrichum malorum*. It also differs in spore shape from *S. lyococcon* and *S. thümenii*, the two species listed by Oudemans (9) as occurring on apple fruit. It is, therefore, considered to be a new species, for which the name *Sporotrichum carpogenum* is proposed.

When inoculated into ripe Jonathan apples, this species was found to cause a dark brown, rather firm decay, at about 20° C. and at cold-storage

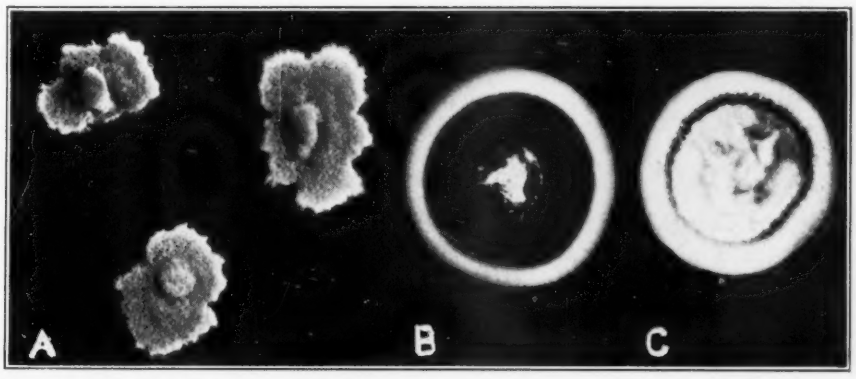

FIG. 2. A, Culture of *Sporotrichum carpogenum;* B, *S. malorum,* dark form; C, *S. malorum,* light form. All the same age on 2 per cent dextrose-potato agar.

temperatures. The rot lesions so produced are not markedly different from those produced by *Sporotrichum malorum* but were found to develop slightly slower at both temperatures. Spores and mycelium of the fungus were abundant in the host tissues, and reisolations yielded the original form in every case.

Botrytis mali, n. sp. This form is differentiated from *B. cinerea* by the production of very small sclerotia, by its shorter, more frequently branched conidiophores, and by its ovate to ellipsoidal conidia, produced on sterigmata which approximate half the length of the spores. By means of inoculation experiments on ripe apples, it was found to be capable of producing a rapid decay, which is of the same type as that produced by *B. cinerea,* but which develops slightly slower at all temperatures. The lenticel spotting noted for *B. cinerea* (3) apparently does not develop in lesions produced by the new form.

On 2 per cent dextrose-potato agar, the fungus grows somewhat slower than *Botrytis cinerea,* the colonies attaining a diameter of 75 mm. after 4 days' growth at a temperature of 25° C. The colonies are white at first, but soon become smoky gray from the production of numerous conidiophores and conidia (Fig. 3). Sclerotia begin to form in about a week and the colonies then gradually turn a light brown. The following is a technical description of the fungus as it develops on 2 per cent dextrose-potato agar.

Mycelium hyaline, septate, variable in diameter, the branches sometimes constricted slightly at the base; conidiophores erect, arising directly from the mycelium or sometimes from sclerotia, septate, simple or with numerous short side branches ending in globose swollen structures that bear conidia on small sterigmata; sterigmata attaining half the length of the spores or shorter; conidia continuous, hyaline, when single, but smoky gray in mass, standing close together to form dense heads; conidia ovate to ellipsoidal, usually finely apiculate at the base, 10–18 x 6.8–10.5 μ, average 11–14 x 7–9 μ; sclerotia white at first, becoming black at maturity, oval on top, flattened at the base, usually from 1 to 2 mm. in width but ranging from mere specks to 3 mm. (Fig. 3, B), germinating to produce conidiophores or to form a mycelium; attachment organs not formed in contact with Petri dish.

Cladosporium malorum, n. sp. Colonies on 2 per cent dextrose-potato agar, and Czapek's-solution agar, a Roman green (Ridgway's color standards), dense, with fluffy surfaces (Fig. 1, B), attaining a diameter of 60–65 mm. in 10 days when grown at 25° C. Hyphae are hyaline at first, becoming light olive; from 2 to 5 μ in diameter, branched, septate at short intervals, not constricted at the septa. The conidiophores are simple, septate, colored as the hyphae, and short. The conidia are light olive, oblong-cylindric with rounded ends, smooth-wall, mostly continuous, many 1-septate,

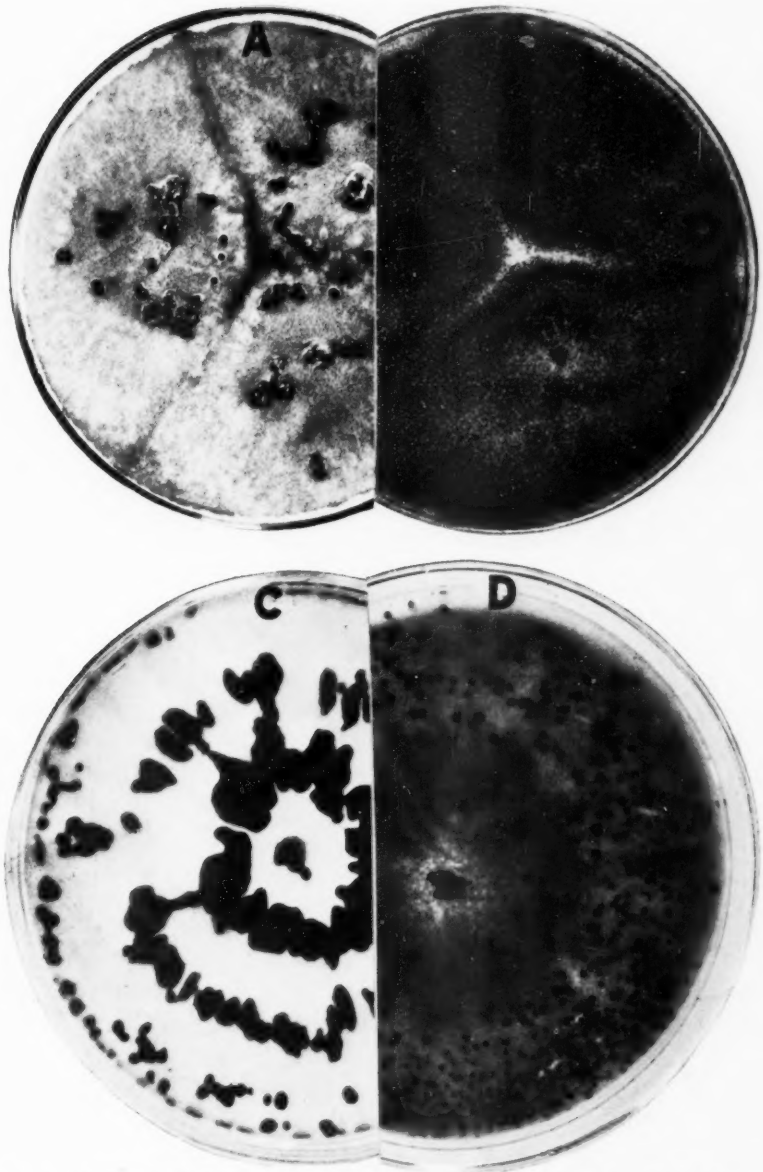

FIG. 3. Cultures of *Botrytis mali* and *B. cinerea* from both upper and lower surfaces. A, *B. cinerea*, upper surface with moderate sporulation; B, *B. mali*, upper surface with heavy sporulation; C, *B. cinerea*, lower surface showing large sclerotia; D, *B. mali*, lower surface showing numerous small sclerotia.

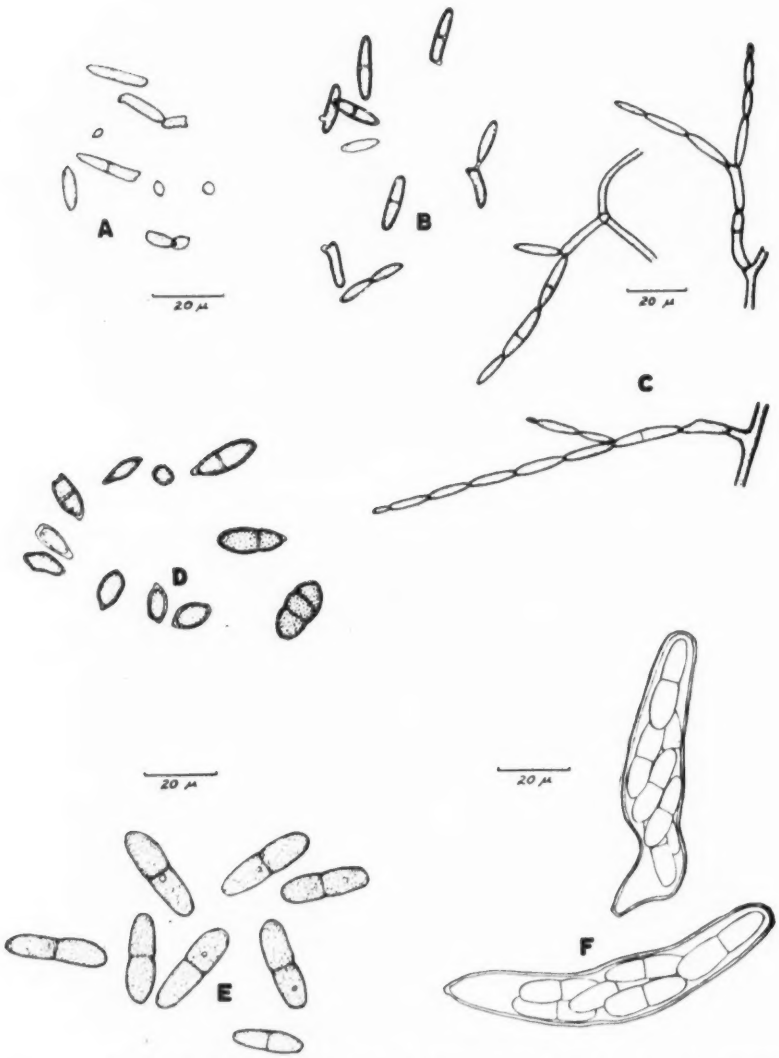

FIG. 4. Drawings of Cladosporium-like forms from apple rots. A, Conidia of *Hormodendron cladosporioides;* B, mature conidia of *C. malorum;* C, young conidial chains of the same species; D, conidia of *Mycosphaerella tulasnei;* E, isolated ascospores of *M. tulasnei;* F, two asci of this species containing ascospores.

produced in long branched chains. Each conidium arises as a bud on that immediately behind, so that the youngest conidium is at the distal end of the chain. The spores are 10–21 x 3–5 μ, the average being 14.7 by 3.6 μ (on the basis of 100 spores), (Fig. 4, B, C).

Inoculation experiments showed that the fungus is capable of producing decay of ripe apples. At 20–25° C., dark brown lesions 20–30 mm. in diameter were formed at the end of 14 days. The rotted areas are fairly firm, and the affected tissue is light brown, rather dry and spongy. The original form was recovered from tissue plantings made from the advancing edge of such lesions. At 0° C., the fungus develops very feebly on the apple, producing small spots at the points of inoculation, which do not spread to cause decay. Although inoculated apples were incubated for 5 months at this temperature, these spot lesions did not advance beyond 10 mm. in diameter. The fungus is able to grow on culture media at this temperature, however, producing colonies that attain 35 mm. in diameter at the end of 2 months. Such colonies are loosely fluffy and produce very few spores.

MYCOSPHAERELLA TULASNEI AND CLADOSPORIUM SPECIES

In the present study fungi of the Cladosporium-Hormodendron type were repeatedly isolated from small, brown to black, firm, shallow lesions developing in apples in cold storage. The lesions did not exceed 20 mm. in diameter and occurred invariably at breaks or injuries to the skin of the fruit, such as bruises, stem punctures, insect injuries, or scalded areas. The morphology of the various isolations was studied from colonies developed from single spores. Eleven strains or species were recognized, possessing fairly constant characters when grown more than a year on culture media. Of these only three were found capable of causing decay when inoculated into apples, as follows:

1. *Cladosporium malorum* described above as a new species.
2. *Cladosporium herbarum* Lk. Colonies very dense with scant development of aerial hyphae, conidia variable in size and shape, 0–3 septate, with roughened walls in branched chains (Fig. 4, D).
3. *Hormodendron cladosporioides*. Similar to No. 2, but conidia smaller; 0–2 septate, smooth (Fig. 4, A).

Although types 2 and 3 have been considered by various writers as stages of the same fungus, it should be emphasized that with frequent transfers and growth at varying temperatures for a period of 3 years, they retained their specific characters. No. 2 developed a perithecial stage, identified as

Mycosphaerella tulasnei Jane., but No. 3 could never be induced to form perithecia.

In 1894 Janczewski (5) described one form of *Cladosporium herbarum* as belonging to the life cycle of *Mycosphaerella tulasnei*, but no confirmation of an ascigerous stage of *C. herbarum*, since that first report, has been found in any of the literature studied. This early discovery of Janczewski appears to have been either ignored or received with some doubt by modern writers. The definite confirmation of this report should be of interest.

Mature perithecia were produced in small numbers on culture media such as corn-meal agar or potato-dextrose agar, when held at low temperatures for long periods, but more abundantly on sterilized wheat leaves inoculated with spores of the Cladosporium stage and incubated at 8–10° C. for a period of 6 months. These perithecia agreed, in general, with those originally described by Janczewski.

Single ascospores were isolated and grown in culture, with the resulting colonies similar in all respects to the original colonies grown from single conidia. There seems little doubt that this form isolated from the apple is the same as the species described by Janczewski, although he reported somewhat larger perithecia and larger asci. At the end of 6 months the perithecia on sterilized wheat leaves were black, with thick wall, typically broadly flask-shape with a short neck, and partially embedded in the leaf tissue, 150–250 μ high by 100–150 wide. The asci are cylindrical, slightly tapering at the ends, 80–120 x 15–20 μ; ascospores hyaline, bicellular, 18–28 x 6–8.5 μ (Fig. 4, E, F).

ADDITIONAL NOTES

In 1928 Newton (8) described a new species of Pleospora from decaying apples in the Pacific Northwest and named it *Pleospora mali* Newton. The conidial stage was classified as a Stemphylium. *Pleospora mali* Newton should not be confused with *P. mali* Hesler, an entirely different species, recently described as the perfect stage of *Hendersonia mali* Thüm (4). Since the published description of *P. mali* Hesler antedates a few months the account by Newton, the specific name given by Newton to the apple-rotting Pleospora must be considered invalid. *Pleospora fructicola* (Newton) Ruehle is proposed for the species described by Newton.

A fungus of the Microdiplodia type has been obtained several times from small, dark brown, firm lesions (20 mm. or less) occurring at punctures. Cultures on potato-dextrose agar fruit readily, especially at temperatures of 10–15° C. The colonies at first are white, but soon become olive brown, with white advancing border. The aerial mycelium, which is white at first, becomes dark brown with age and is partly united into small compact

strands consisting usually of 4–6 hyphae. The pycnidia are scattered, or sometimes in groups of 2 or 3, subglobose, nonostiolate, dark brown, 100–230 μ in diameter with thin, easily ruptured walls 2–3 cells thick. The pycnospores are light to dark brown, usually 1-septate (occasionally 2–3) oval to oblong with rounded ends, smooth wall and 9.5–15.5 x 6.9–9.5 μ. Inoculation of ripe Jonathan apples at laboratory temperatures gave brown, firm, shallow lesions which reached a diameter of 25 mm. or less in 60 days. No pycnids were formed on the fruit lesions. No records of the previous occurrence of a Microdiplodia causing apple decay have been found.

A species of Epicoccum, agreeing closely with *E. granulatum* Penz., was obtained from a dark rot on a Rome Beauty apple. When inoculated into ripe Jonathan apples it produced a firm, rather dry, reddish brown rot in which cavities were sometimes formed, both in cold storage and at 20° C. In cold storage the lesions were 10–35 mm. in diameter after 5 months, while at the higher temperature they were 20–25 mm. in diameter after 2 months. The affected tissue was sometimes reddish brown and dry, but in larger lesions it was soft, moist and bright red. No species of Epicoccum have been previously reported as capable of causing decay in apples.

Several fungi have been isolated from apples which have not been assigned to definite species. These appear to be distinct from species of these genera previously recorded as affecting apples, but it has not seemed advisable to give them specific rank until they can be given a more detailed study. The isolations falling in this class are:

Phoma Nos. 1 and 2.
Coniothyrium Nos. 1 and 2.
Oospora sp.
Alternaria Nos. 3, 4, and 5.
Fusarium Nos. 1 and 2.
Ramularia No. 2.

FLORIDA CITRUS EXPERIMENT STATION,
LAKE ALFRED, FLA.

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A COMPARISON OF PSEUDOMONAS PRUNICOLA WITH A CANKER-PRODUCING BACTERIUM OF STONE-FRUIT TREES IN CALIFORNIA

E. E. WILSON[1]

INTRODUCTION

In 1928 Wormald (8) described a wilt disease of the green shoots of plums in England and later (9) presented proof that this disease is caused by a bacterium, to which he gave the name *Pseudomonas prunicola*, n. sp. The organism differed in major characteristics from *Bacillus spongiosus* Ader. and Ruh. and from *Bacterium* (*Pseudomonas*) *pruni* E. F. Smith. It also differed in certain details from *Pseudomonas cerasi* Grif., as described by Griffin (6) and Barss (2).

The present paper describes the results of a comparative study of *Pseudomonas prunicola* Wormald, with an organism found commonly associated with a gummosis of stone-fruit trees in California. The latter organism possesses distinct pathogenic abilities but does not possess the chromogenic ability attributed to *Ps. cerasi*.. Due to this difference and the fact that a second type of organism has been found that does possess the chromogenic ability, the writer prefers, for the present at least, to designate his organism as 357. These points will be discussed more in detail in the body of the paper.

Dr. Wormald kindly furnished the writer with a culture of *Pseudomonas prunicola*. The culture of 357 was obtained from blighted apricots buds during the winter of 1929–30.

MORPHOLOGICAL AND STAINING STUDIES

Both *Pseudomonas cerasi* (6) and *Ps. prunicola* (9) are described as rods motile by 1 to 3 polar flagella and commonly occurring in pairs. The former is reported to be from 1.5 to 2.4 μ long by 0.5 to 0.84 μ wide; the latter, 0.9 to 2.5 μ long by 0.3 to 0.5 μ wide.

In the present study 357 and *Pseudomonas prunicola* were grown comparatively on beef-extract agar. Smears taken at 48 and 120 hours were stained with safranin. Table 1 presents the results of measurements by two persons. Only those cells were measured that showed neither a constriction nor a transverse separation. In this way, errors arising from measurements of cells in the process of division or of connected cells were minimized.

The data of table 1 show no consistent difference between the size of the two organisms. The differences obtained by the two observers of measure-

[1] The writer wishes to acknowledge his indebtedness to Professor R. E. Smith for advice and criticism.

ments of cells in the same smears were probably incidental to their abilities to estimate fractions of a space on the ocular micrometer.

Wormald (9) states that *Pseudomonas prunicola* may at times become linked together in long chains, a characteristic which has also been noted for 357. The phenomenon occurred in beef-extract agar cultures of 357, which had been growing at room temperature for 120 hours. When such chains were stained with methylene blue, it was seen that the organisms were held together by a common hyaline sheath.

Wormald (9) found that *Pseudomonas prunicola* was slightly Gram positive if he used either the method of Eyre or those methods outlined in the S. A. B. Manual. In the present work, smears of 357 and *Ps. prunicola* grown on beef-extract agar were taken at intervals and stained by the aniline-gentian-violet method outlined in the S. A. B. Manual. Although both organisms retained some of the gentian violet, the method was deemed unsatisfactory, inasmuch as the stain was precipitated as particles over and around the cells. When the procedure was modified by washing the smears in water between steps, this precipitation did not occur, nor did the cells retain the stain.

TABLE 1.—*Measurements by two persons of 357 and Pseudomonas prunicola grown for different lengths of time on beef-extract agar*

Organism and age in culture	Observer No. 1			Observer No. 2		
	Extremes of length	Extremes of width	Average length and width	Extremes of length	Extremes of width	Average length and width
	Microns	Microns	Microns	Microns	Microns	Microns
357:						
48 hours	1.1–2.5	0.5–0.7	1.8 × 0.7	1.1–2.8	0.5–0.7	1.6 × 0.7
120 hours	0.9–2.3	0.5–0.7	1.7 × 0.5	0.8–2.8	0.5–0.7	2.2 × 0.7
Ps. prunicola:						
48 hours	1.1–2.8	0.5–0.7	2.1 × 0.7	1.1–3.2	0.5–0.7	1.9 × 0.5
120 hours	1.1–2.8	0.5–0.7	1.8 × 0.5	1.3–2.8	0.5–0.7	1.8 × 0.6

CULTURAL STUDIES

Pigment production.—Wormald (9) did not consider *Pseudomonas prunicola* to be identical with *Ps. cerasi* inasmuch as the latter was reported to produce a green pigment, while the former produced a lemon-yellow pigment, which diffused through the medium, leaving the mass of organisms uncolored.

The various workers on the gummosis of stone fruit are not definite as to the chromogenesis of the organisms they found causing the disease. Griffin (6) (see also Barss, 2) states that the greening of a number of

media was a constant characteristic of *Pseudomonas cerasi*. Barss (2) did not report results of his cultural studies but was evidently in accord with Griffin on this point. Barrett (1) does not give the cultural characteristics of the organism he found to be the cause of apricot and plum gummosis in California. Goldsworthy (5), working in California, reported a fluorescent and a nonfluorescent type capable of producing gummosis. The fluorescent characters was not constant. While the two types were indistinguishable by cultural tests he was able to show that they produced specific agglutinins in experimental rabbits. Smith and Fawcett (7) state that their isolation of *Ps. cerasi* differed in its utilization of lactose and maltose from that reported by Griffin but do not mention pigment production.

The organism (357), which has been found in the present study to be most common in gummosis cankers, produced a yellow rather than a green discoloration in beef-extract media. On potato-glucose agar only a slight yellow tinge appeared after the organism had been growing for a number of days. This characteristic on potato glucose distinguished 357 from a second type (506), which has been found less frequently. On this medium, and to a lesser degree on beef-extract media, the second type produced a brilliant yellowish green pigment (approaching an apple green by reflected light; Ridgway[2]) that possessed fluorescent qualities. This type has produced gumming cankers upon being inoculated into plum and apricot trees and has been recovered from such cankers several months later.

It is beyond the scope of this paper to deal with the importance of the two types in the general problem of gummosis. A more exhaustive study is necessary before this phase can be properly evaluated. Their existence has been mentioned in order to show why the writer is unwilling to designate the more common one (357) as *Pseudomonas cerasi*. As shown later, 357 also differed from *Ps. cerasi* in its utilization of lactose and maltose.

In a series of tests 357 and *Pseudomonas prunicola* were similar in type of growth and discoloration of beef-extract media. After 7 to 10 days the medium was changed to a lemon-chrome or lemon-yellow (Ridgway[3]).

In a medium composed of 0.5 gm. ammonium dihydrogen phosphate, 0.2 gm. potassium chloride, 0.01 gm. calcium chloride, and 10 gms. succinic acid per liter and adjusted to pH 7.0 with sodium hydroxide, both organisms produced a characteristic greenish yellow color. While the color did not possess marked fluorescent qualities by daylight examination, it would probably fall into this category.

[2] Ridgway, Robert. Color standards and color nomenclature. Washington, D. C. 1912.

[3] *Op. cit.*

Pseudomonas prunicola differed from 357 in these tests only in becoming somewhat flocculent by the 6th day, while 357 remained evenly suspended.

Utilization of nitrates.—Following the procedure outlined in the S. A. B. Manual, a study was made of the growth of 357 and *Pseudomonas prunicola* on media containing nitrates. A preliminary test had failed to show the reduction of nitrates to nitrites. The next step was to grow the organisms on four different media as follows: (a) potassium nitrate-beef-extract broth; (b) beef-extract broth without the addition of potassium nitrate; (c) a synthetic medium containing potassium nitrate, dipotassium phosphate, calcium chloride, and glucose; and (d) a beef-extract broth containing 2 parts per million of potassium nitrite. Using the reagents recommended in the Manual, "a" and "b" were tested for ammonia, "c" was tested for both ammonia and nitrites, and "d" was tested for nitrites.

The tests gave no indication that either organism produced nitrites or ammonia. Upon applying the test for ammonia to a and b, a yellowish color appeared in the bottom of the tubes. This slowly changed to a green, which lasted for 20 to 30 minutes and then disappeared, but no blue color appeared at any time. There was evidence that both organisms were capable of utilizing nitrite, as the test for this substance in d became weaker and weaker until by 96 hours no test could be obtained.

Utilization of various carbon compounds.—*Pseudomonas cerasi* was originally described (Barss, 2) as producing acid on lactose and maltose. This was expressed in terms of the titratable acidity (Fuller Scale). Smith and Fawcett (7), basing their tests on the change in pH, have reported that their isolation of *Ps. cerasi* produced alkali on these sugars.

Wormald (9) found that *Pseudomonas prunicola* was able to grow in "nutrient broth" to which no carbohydrate had been added. The medium under these circumstances became more alkaline. If either glycerin or lactose was added to the above medium the bacteria caused the pH to be shifted towards the alkaline side. If, however, either glucose or sucrose was used the bacteria shifted the pH towards the acid side.

The experience of the writer has been that, if the bacteria produce small amounts of acid from a given carbon source, its presence may be masked by the alkali produced from their action on the peptone, if peptone is used in the medium. Thus, it was found that in beef-extract broth with glycerin as a carbon source, 357 often produced alkali, while in a medium composed of inorganic salts and glycerin, the organism produced acid. Studies of the utilization of carbon compounds were therefore made on both types of media.

Table 2 presents data obtained by growing the two organisms on beef-extract agar with various sugars as carbon sources. The medium was ad-

justed to a reaction of pH 6.5, with brom thymol blue as an indicator. With the exception of the glycerin medium, the two organisms produced very similar changes of pH.

TABLE 2.—*Comparison of change in pH produced by 357 and Pseudomonas prunicola in beef-extract agar containing different carbon sources*

Carbon source	Organism	Change[a] from control after stated time				
		2 days	3 days	5 days	8 days	15 days
Galactose	*Ps. prunicola*	0	+++	+++	+++	+
	357	++	+++	+++	+++	+
Glucose	*Ps. prunicola*	++	+++	+++	+++	−
	357	++	+++	+++	+++	−
Glycerin	*Ps. prunicola*	−	−	0	0	+
	357	0	+	++	++	++
Lactose	*Ps. prunicola*	−	−	−	−−	−−−
	357	−	−	−	−−	−−
Sucrose	*Ps. prunicola*	+	+	++	++	−−
	357	+	0	++	++	−

[a] 0 = No change from pH 6.5; (+) = change towards acid side; (−) = change towards alkaline side.

The general appearance of the two organisms in the agar streaks was strikingly similar. One rather common characteristic was the occurrence of minute pits in the surface of the growth, which resulted in a rather dull appearance (matte-effect). The condition did not always persist but gave place to a smoother and more glistening surface. The growth was rather flat on all of these media; the margins were generally slightly lobed and the lobes, in turn, were irregularly toothed. In the case of the dextrose and glycerin media, the margins tended to be more entire.

In the inorganic medium mentioned under "Pigment production," both organisms shifted the pH towards the alkaline side when the carbon source was either lactose, raffinose, trehalose, or succinic acid. When either glucose or sucrose was used there was an increase in hydrogen-ion concentration. Neither organism appeared to be able to utilize rhamnose to any great extent, as no growth had occurred in this medium after 72 hours. Figure 1 shows graphically the results obtained in the glucose medium. While the curves do not parallel each other at all points, there is a general agreement in their conformation, in that both organisms produced a rather slow in-

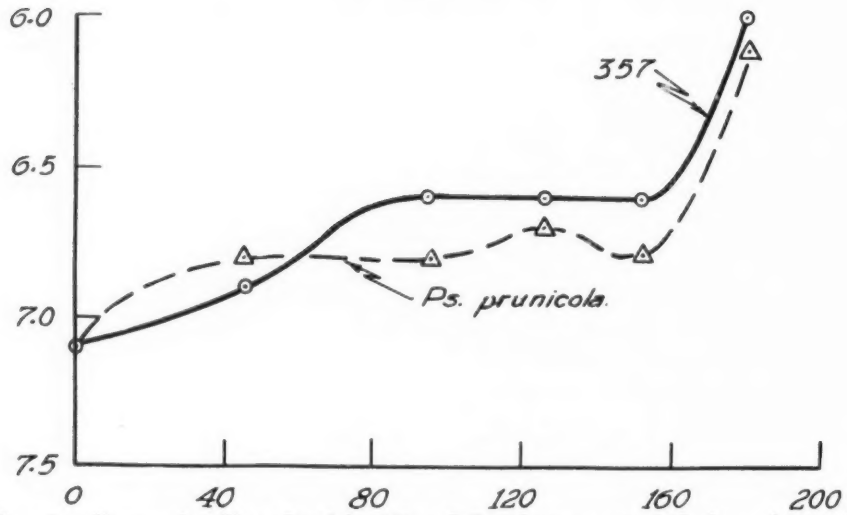

FIG. 1. Changes in pH produced by 357 and *Pseudomonas prunicola* in an inorganic liquid medium containing glucose as a carbon source. Hydrogen ion concentration plotted against time in hours.

crease in hydrogen-ion concentration during the first 148 hours, after which there was a rather sudden increase.

PATHOGENICITY

Pseudomonas cerasi is capable of attacking under natural conditions the leaves, the buds, and limbs of stone fruits (Barrett, 1; Barss, 2, 3, 4). It has not been reported to attack the green shoots in nature but has been found by Smith and Fawcett (7) to produce lesions on the green shoots of a rather wide variety of hosts when inoculated artificially. Even though natural infection of green shoots was possible, the low humidity conditions of the Pacific Coast during the growing season would probably militate against an abundance of this type of injury.

Wormald (9) stated that *Pseudomonas prunicola* produced gumming cankers when inoculated into the limbs of plum but that these cankers never became large enough to girdle the limbs. He (9) further observed that *Pseudomonas prunicola* had never been found associated with naturally occurring cankers on "woody stems" of the plum but mentions a second bacterium that did produce extensive natural cankers. The description of the second bacterium was reserved for a later publication.

Since *Pseudomonas prunicola* and 357 agreed in most of the culture tests used, the two organisms were inoculated into the limbs of plum and cherry trees by means of a hypodermic needle. The small wounds thus made were covered with either vaseline or oiled paper.

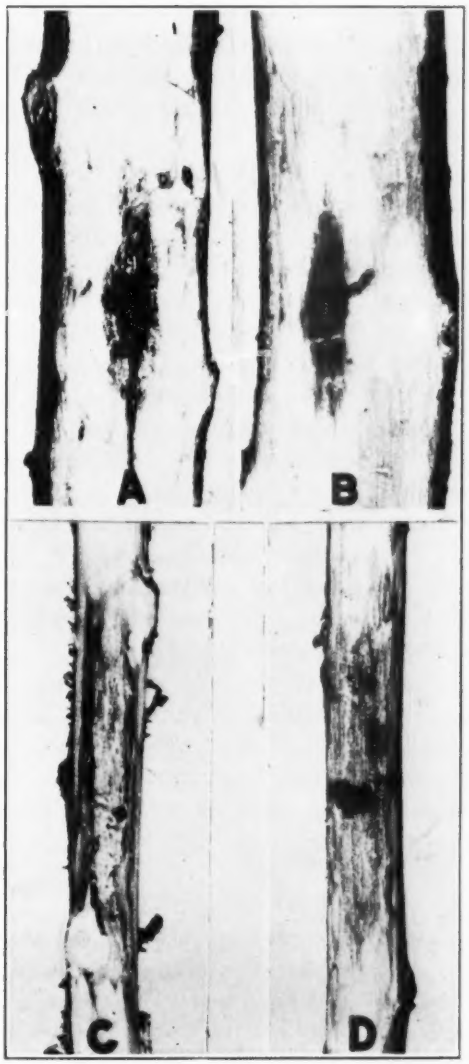

FIG. 2. Comparison of cankers produced by (A) 357 and (B) *Pseudomonas prunicola* on Duarte plum branches and by (C) 357 and (D) *Ps. prunicola* on President plum branches. $\times \frac{2}{3}$.

In a series of 36 inoculations into Duarte plum on January 14, 1931, there was never much evidence of gumming from the points of inoculation, but by February 18 depressions were noticeable around the wounds. Upon cutting into the bark definite cankers were found that were from 1 to 3 inches long and from $\frac{1}{2}$ to 1 inch wide. Both organisms produced cankers similar in all respects (Fig. 2, A, B). It was characteristic for gum to be exuded both from the edge and in advance of the necrotic areas of these cankers when the bark was cut.

On January 15, 1931, 20 inoculations were made into young German Mazzard cherry trees, and 36 inoculations were made into young trees of Grand Duke, President, and Wickson plums. Neither 357 nor *Pseudomonas prunicola* produced cankers over 1 inch long on the cherries. On the plums, however, both organisms had produced by March 3 cankers from 3 to 6 inches long (Fig. 2, C, D). These cankers were similar in all respects. The most rapid progress had been made along the cambium. The edges of the cankers were generally watery or gum-impregnated, while the centers were dark brown. Gum-soaked streaks occurred through the phloem region and would often terminate in gum pockets.

A series of 15 inoculations was made into limbs of Bing cherry on January 15, 1931. By January 25 abundant gum had been exuded from the point of inoculation. When the final observations were made on March 5 it was found that both organisms had produced cankers from 2 to 3 inches long and from $\frac{3}{4}$ to 1 inch wide. Gum pockets were present at times 2 or more inches from the point of inoculation.

Organisms that agreed in all respects with those used for inoculations were recovered from these cankers. It was comparatively easy to obtain a pure culture by removing bits of bark from the edges of the cankers and dropping them into broth tubes. The same was true if such bits of bark were allowed to stand in sterile water for an hour or two, after which platings were made from the water.

SUMMARY AND DISCUSSION

A comparative study has revealed that *Pseudomonas prunicola* resembles very closely an organism, designated in this paper as 357, which produces a gummosis disease of plum and apricot trees in California.

The latter organism differs in certain cultural details from *Ps. cerasi*, the reported cause of gummosis of cherry trees in Oregon.

A second type of organism (506) is noted in California, which resembles *Ps. cerasi* more closely in chromogenesis. It is also capable of producing a gummosis type of canker on plum and apricot. More detailed work is necessary to determine the relationship of the two types to each other and to *Ps. cerasi*.

The work of Goldsworthy (5) is cited as indicating that two very similar organisms may cause gummosis of stone-fruit trees. Since this worker failed to report the cardinal features of his organisms it is impossible to compare them with those found in the present work. It is possible that the organism designated in this paper as 506 is identical with Goldsworthy's fluorescent type. The organism designated 357, however, does not appear to be identical with Goldsworthy's second type, inasmuch as the former produced a fluorescence in certain media, while the latter is reported to be nonfluorescent.

With the information at hand regarding *Ps. cerasi* and according to the general concepts of species limits, Wormald was probably justified in creating a new species. In view of the situation set forth in this paper, however, the writer hesitates to accept it as a new species until further work has been done to prove its relationship to *Ps. cerasi*.

DIVISION OF PLANT PATHOLOGY,
BRANCH OF THE COLLEGE OF AGRICULTURE,
DAVIS, CALIFORNIA.

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CORYNOSE TWIG BLIGHT OF THE AMERICAN BLADDER NUT, STAPHYLEA TRIFOLIA

W. H. Davis

For the past three growing seasons, blighted twigs bearing fruiting bodies of a Coryneum have been observed on shrubs of the American bladder nut, *Staphylea trifolia* L. Since no reference to this disease was found in our best host indexes and available literature, an investigation was begun in the spring of 1930 with a view to answering the following questions:

1. Is this Coryneum the pathogen causing the twig blight?
2. How prevalent and injurious is the disease?
3. When and where does infection occur?
4. What is the Latin binomial of the fungus?
5. Is there an ascogenous stage?
6. From a study of the life history of the fungus, what methods of its control can be recommended?

SYMPTOMS AND SEASONAL OCCURRENCE

The first symptoms of the disease resulting from new infection during the current year were observed in August. These symptoms were a dying of the tips of the youngest twigs or the ashen gray color of the nodes. Later, however, lower nodes on infected twigs assumed a cartridge buff, the young buds died or failed to appear, and areas at internodes often assumed a cinnamon rufous hue. The infection sometimes advanced to a node where the Roman green color of the healthy bark could be easily differentiated from the buff of the infected area (Fig. 1, I). At this point, the tissues in the diseased area often contracted and wrinkled so as to leave a raised girdle around the twig between healthy and diseased tissues. The girdle appeared as a collar varying from a fine line, 0.5 mm. to 5 mm. in width (Fig. 1, I, 3). Furthermore, the organism, which generally entered at the nodes, advanced along both the longitudinal and radial axes of the stem but its longitudinal advance generally was halted at branches or at the beginning of the current year's growth. Furthermore, the organism advanced from younger twigs into older ones that often died during or at the close of the growing season. The disease appeared to advance during early winter, when more diseased twigs were observed than at the close of the growing season. As a final result, 30 per cent of the current year's twig growth was killed on some shrubs.

In 1930, the fruiting bodies or acervuli were first observed on newly infected wood during September. Acervuli formed around that portion of the twig receiving the initial infection or near the node. These acervuli,

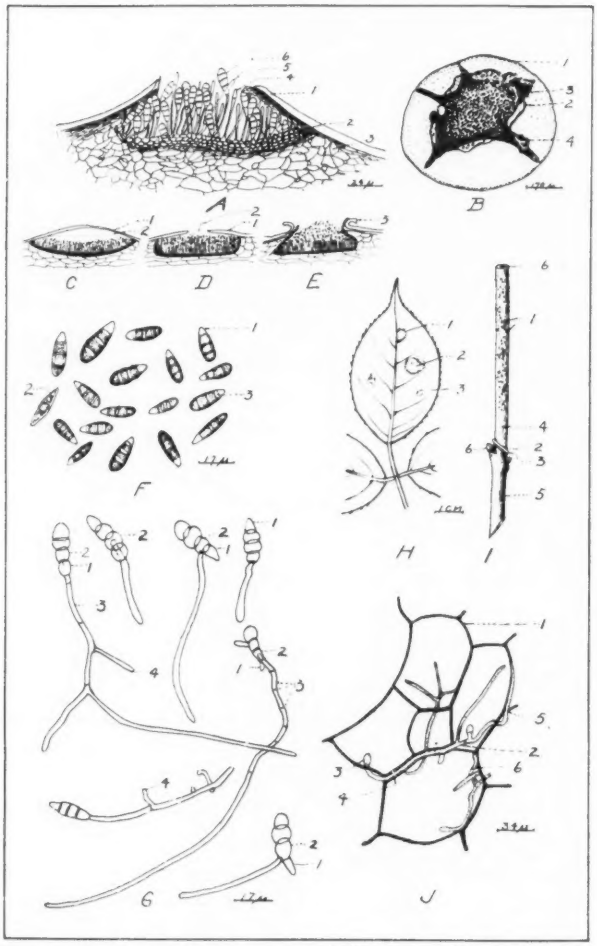

FIG. 1. *Coryneum microstictum* var. *staphyleae*. A. Vertical section of an acervulus; 1, epidermis of the host; 2, sclerotial cells; 3, cortical tissue; 4, immature conidium; 5, conidiophore about to form a conidium; 6, mature conidium. B. Acervulus on bark of host; 1, margin of the brown, "oily"-appearing bark; 2, recurved epidermis; 3, conidia; 4, rift in the bark. C, D, E. Vertical sections of acervuli; C, saucer-shape or flat before rupture of the epidermis; D, with epidermis ruptured; E, the long conidiophores projecting through the rupture make the acervulus appear conical. F. Conidia; 1, 2, hyaline pedicellate cell; 3, septum. G. Conidia germinating in water; pedicillate cell, 1, not germinating, but with cell next to it; 2, germinating and pushing it aside; 3, cross walls; 4, branches. H. Leaflets; 3 weeks after inoculation; 1, 2, 3, circular lesions. I. An infected twig; 1, acervuli; 2, limit of diseased bark; 3, collar between healthy, 5, and diseased bark, 2; 4, wrinkles in the diseased bark; 6, node. J. Pith cells from diseased twig hand-sectioned and stained with lacto-phenol containing acid green: 1, cells; 2, mycelium; 3, haustorium; 4, intercellular hypha; 5, hypha penetrating a cell wall; 6, intracellular hyphae.

when observed by aid of the hand lens, appeared as black, slightly raised, circular areas averaging 0.5 mm. in diameter, arranged singly but numerous and in close proximity or gregarious (Fig. 1, I). Bark peeled from 1 cm. along the longitudinal axis of a diseased twig contained 158 acervuli.

Acervuli that had remained in diseased twigs during the winter often appeared ashen gray and bore longitudinal rifts through the center (Fig. 1, B, C, D, E). However, some acervuli raised the epidermis of the host and might have been mistaken for the beaks of perithecia. Sometimes, small areas around the rifts of acervuli and the surface of the twig were an ashen gray, while the acervuli were of a red (tawny) color, which often disappeared as the growing season advanced. This change of color was most noticeable in old stems that had become infected by the fungus advancing from younger twigs.

Typical springtime symptoms on twigs of the previous year's growth were ashen gray bark with shrunken surface in which were slightly raised, black, gregarious acervuli. Matured acervuli were somewhat elliptical in shape, with longitudinal slits through which the honey-color spore masses could be observed.

Acervuli were also observed on infected, dried wood exposed by the shrinking and withdrawal of the diseased bark.

Conidia were formed during late summer and early autumn in the majority of the acervuli examined. Here they passed the winter and during the springtime (April and May), the epidermis of the host covering the acervuli cracked and the conidia were exposed to the air for dissemination. Few of the conidia, formed in the autumn of 1929 and 1930, germinated during those autumns but 90 per cent germinated during the spring (March, April, and May) of 1930 and 1931 (Fig. 1, G).

PATHOLOGICAL ANATOMY

Hyphae of the specific Coryneum involved in this study were located in medulla, xylem, and cortex of infected stems examined during the late spring and summer. Hand sections showed that hyphae in the pith were both inter- and intracellular (Fig. 1, J) and that they often traversed the medullary rays. Hyphae advanced nearly 8 inches in one inoculated twig during 8 months of incubation. Hyphae also were located throughout the pith of "water sprouts" that had grown 2 feet in length during the current year. This, however, might have been due to hyphae advancing from several and not from one nodal infection.

During the winter of 1930–1931, experiments were performed to determine whether mycelium remained viable within wood of the American bladder nut or only the conidia survived the northern-winter conditions.

Young twigs infected during 1930 and infected twigs of various ages were removed from shrubs during November, December, January, February, and March. Each of the five lots were disinfected with formaldehyde and washed in sterile water and portions of tissues removed from pith, wood, and bark of both the diseased and the healthy wood near, remote, and at the last visible point of infection. These portions were transferred to potato-dextrose agar under aseptic conditions and incubated at 20° C.

The results showed that the hyphae remained viable in nearly all the diseased twigs during November; in 10 per cent of the old twigs during December and January. Viable hyphae were also cultured from old twigs collected in February and March.

Thus, the hyphae of the organism remained viable, during the winter of 1930, in the older twigs of the bladder nut but not in young ones including the water sprouts.

ISOLATING AND CULTURING THE FUNGUS

Single conidia were isolated on April 25 and monosporous cultures were incubated at 22° C. on potato-dextrose agar in Petri dishes and test tubes. Furthermore, transfers of the fungus were made on steamed rolled oats and yellow corn meal in 500 cc. flasks.

The spores germinated readily and the fungus grew rapidly on each of these three media. Acervuli containing viable conidia were observed in the flask cultures on corn meal in 18 days and on rolled oats in 32 days after the transfers were made.

At first, the mycelium growing on potato-dextrose agar was a grayish white; after 3 to 5 days it changed to a light gray, then became a flesh color, rufous or chestnut brown, but finally became a dark sepia or black when old.

The growth was somewhat strict and rugose and formed a pellicle on which were acervuli. Acervuli were most numerous on the sides of the flask cultures facing the sunlight. The fungus grew faster and sporulated sooner on steamed corn meal, which was considered the best culture medium tested. The culture on steamed rolled oats, however, remained viable for 9 months, the longest period observed.

On May 14, the buds on *Staphylea trifolia* had formed young leafy stems 4 to 6 inches long. Young twigs and leaves in this condition were inoculated with a composite suspension of spores removed from three monosporous cultures, together with a small amount of mycelium. The inoculations were performed on shrubs outdoors and the inoculum placed on (a) punctured stems at leaf axils; (b) unpunctured stems at leaf axils; (c) punctured internodes and twig tips; (d) unpunctured internodes and twig

tips; (e) leaves, punctured and nonpunctured; (f) checks, punctured and nonpunctured stems and noninoculated leaves.

The surface of the twigs was sterilized with an 8 per cent aqueous solution of formaldehyde, washed with sterile water, inoculated, atomized with sterile distilled water, and then covered with oiled bags containing a wad of sterile, wet cotton. After 2 weeks, the bags were removed. Furthermore, to determine the time or season during which infection occurred, inoculations were made later in the season, when a majority of the spores were discharged from the host and the plant tissues inoculated were more matured. The date chosen was July 22.

During the following November, acervuli in some inoculated twigs contained conidia sufficiently matured for recognition of the fungus.

From the results recorded in table 1, it is to be noted:

1. The fungus was pathogenic.

2. Primary twig infection occurred in immature wood, at the leaf axils or nodes and at the tips or in meristematic tissues.

3. Infection did not occur at the internodes, even when they were punctured. Further observations showed that the inoculating wounds were generally healed without the symptoms of the disease appearing during the current year.

4. Leaves seldom, if ever, became infected while attached to the plant.

5. Puncturing was not necessary for infection.

6. Meristematic stem tissues, under favorable conditions, were most susceptible to infection.

Furthermore, incised leaves also were inoculated. The surface of the leaflets was sterilized and washed in sterile distilled water, and the leaves placed in Coplin breeding jars, lined with damp filter paper. The inoculum consisted of both spores and mycelium removed from each of three flask cultures and mixed with sterile distilled water. This inoculum was spread on the leaflets as follows: upper and under sides; punctured and nonpunctured surfaces; and at midrib, base, tip, and margin. The inoculated leaflets, together with the checks, were incubated at 25° C. After 2 weeks, small circular ash-color lesions, bordered by wine-color margins, appeared on two of the inoculated areas (Fig. 1, H). After 4 weeks, some of the inoculated leaves turned a brownish black and rotted, while the checks were intact but chlorotic. In no case were acervuli or conidia observed on these inoculated leaves. This experiment was repeated. Only growing twigs were substituted for the leaves and their cut surfaces were set in sterile, distilled water. Six weeks after inoculation, acervuli and conidia formed in the bark of the inoculated twigs. These two experiments, together with field observations, seemed to give evidence that twigs more than leaves favor the entrance and development of the parasite. This was

TABLE 1.—*Tabulated results for the inoculations of Staphylea trifolia with Coryneum microstictum var. staphyleae*

Part inoculated	5–14–30			5–20–30			7–22–30		
	Number inoculated	Infection		Number inoculated	Infection		Number inoculated	Infection	
		Plus	Minus		Plus	Minus		Plus	Minus[d]
Internodes of punctured stems	4	0	4	7	0	7	15	0	15
Internodes of nonpunctured	4	0	4	7	0	7	15	0	15
Tips of twigs, punctured	4	3	1	5	5	0	15	0[b]	14
Tips of twigs, nonpunctured	4	3	1	5	4	[b]	15	1[c]	14
Punctured leaf axils	4	4	0	8	7	1	15	0	15
Nonpunctured leaf axils	4	3	1	8	6	2	15	0	15
Punctured leaflets	6	1[a]	5	0	0	0	0	0	0
Nonpunctured leaflets	6	0	6	0	0	0	0	0	0
Stems and leaflets; checks	0	[b]	5	2	0	2	2	0	2

[a] No sporulation was observed.

[b] Specimen lost.

[c] Latent infection advanced upward from a diseased twig.

[d] Infections may appear after incubating the fungus for more than 1 year.

probably due to the large amount of pith in stems for $\frac{4}{5}$ of the 5 mm. diameter of young twigs is occupied by pith (Fig. 1, J). Here, the fungus seemed to meet with very little resistance in advancing along the longitudinal axis and obtained food sufficient for its healthy growth, since the pith cells were filled early in the season with stored nutrients that were apparently available to the hyphae.

The hyphae overwintered in the nodes and, in the spring, advanced into the buds or downward into living parts of the stem. The cells of the epidermis were seldom invaded but appeared in hand sections as a somewhat homogeneous layer above acervuli. During the growing season, however, hyphae seemed to advance through all parts of young twigs once they were infected.

MORPHOLOGY

Conidia. Over 85 per cent of the conidia observed were clavate or pyriform, while others were fusiform or elliptical. Ninety-eight per cent of the matured conidia were triseptate and few or no spore-wall constrictions were noticeable in fresh materials (Fig. 1, F). The color of the three upper cells was honey yellow or ochre yellow, while the lower or pedicellate cell was subhyaline, and, being sticky when wet, aided in anchoring the conidium (Fig. 1, F and I). Measurements of 100 conidia, removed from twigs in spring condition and mounted in water, follow: limits of variation, 5.1–6.8 × 17–23 μ. Standard, 6 × 19 μ. Few germinated in the autumn but 98 per cent germinated in the spring. One collection of April 24, stored in a living room until November 25 of the same year, germinated 40 per cent. Conidia placed in tap water at 22° C. formed germ tubes that, in turn, formed new conidia after 5 days' incubation.

Conidia in acervuli were borne acrogenously, singly on pedicels which were hyaline, filiform, and averaged 1.3 × 19 μ. However, some were observed as long as 30 μ (40 measured). The conidiophores in acervuli originated from a layer of selerotia-like, polyhedral cells which overlaid 2 to 4 other layers. These cells became a Bismarck brown with age and formed a sclerotial mass; acervuli varied in shape from discoid to conical or pseudoperithecial (Fig. 1, C, D, E). Mature, nonerumpent acervuli, when measured at the surface of the host, averaged 0.6 mm. in diameter, while acervuli, in hand sections, varied at the base from 0.6 to 1 mm. in diameter and, from the unbroken epidermis of the host to the base, acervuli averaged 350 μ (Fig. 1, A).

Although the fungus was cultured for 7 months on three media of varying acidities and on twigs of the host, no perfect or ascogenous stage was observed. None of the old twigs of the host bore an ascomycete which, when cultured, produced a Coryneum. The fungus could overwinter as

perennial mycelium or as viable conidia in the host; therefore, an ascogenous stage would not be necessary for the completion of its annual life cycle.

TAXONOMY

At the present time fungi are classified according to their physiological, as well as morphological, characteristics. So, to determine definitely the Latin binomial of an unidentified form, both of these characteristics should be known. The literature at hand on the genus Coryneum yields very little information regarding its physiology, since most of the species have been established on morphological characters or according to the host from which the specimen was collected. Very few hosts other than the peach have been inoculated with species within the genus. Furthermore, descriptions of species are often inaccurate and incomplete.

Considering the fungus entirely from a morphological standpoint, it compares most favorably with *Coryneum microstictum* B. & Br., as reported by Saccardo[1] and Allescher.[2] The conidia are subpyriform, $6 \times 18\ \mu$ (Saccardo, $5–6.5 \times 15–17\ \mu$); apex, obtuse; 4 loculi, the lowermost one subhyaline, those above, honey color; conidiophores, filiform and hyaline.

These differences, however, were noted; stroma present, not obsolete; conidiophores, $1.3 \times 19\ \mu$, not $1.5 \times 20–25\ \mu$. Yet, a few were $30\ \mu$ in length, but they bore no viable conidia; Staphylea, an unreported host.

Until more information is available regarding the physiology and parasitism of the genus Coryneum the following classification for this parasite is suggested: *Coryneum microstictum* B. & Br. var., *staphyleae*. Host, *Staphylea trifolia* L., and the disease commonly known as Corynose twig blight.

CONTROLS

No definite spraying and dusting program was attempted. Smith[3] recommended a 20–20–200 Bordeaux spray for controlling *Coryneum beyerinkii* Oud., which parasitized peach trees in California. This should be applied in the fall and, again, just before the buds open in the spring.

Just as the bud scales of Staphylea opened in the spring under New England conditions, the conidia were disseminated from the old infected twigs and germinated best. So it would seem that any suitable spray applied at this time would be most effective in killing these conidia. Since initial infection takes place in meristematic tissues the spray, to be most effective, should be placed on these tissues. However, if the twigs bearing

[1] Saccardo, P. A. Sylloge fungorum. 3: 774–775. 1884.

[2] Allescher, A. Fungi imperfecti. In Rabenhorst Krypt. Flora. Abt. 7: 640–641. 1903.

[3] Smith, R. E. California peach blight. Calif. Agr. Exp. Sta. Bul. 191. 1907.

viable conidia and perennial mycelium were pruned and burned, preferably in the autumn, the greatest source of infection would be removed, and this would seem to be the simplest and most effective control method.

SUMMARY

1. The American bladder nut was parasitized by a Coryneum, which, in one case, caused death to $\frac{1}{3}$ of the current year's twig growth.

2. The disease was prevalent in the Connecticut Valley of Massachusetts.

3. The greatest injury occurred in the young twigs as a twig blight.

4. Acervuli, containing viable spores, were formed in the autumn, overwintered in twigs, and disseminated their viable conidia in the spring.

5. Inoculation experiments showed that infection occurred at the nodes and tips of the meristematic stem tissue during the spring.

6. Mycelium of the Coryneum remained viable in some of the old stems, on shrubs, during the winter of 1930–1931.

7. *Coryneum microstictum* B. & Br. var. *staphyleae* is suggested as a Latin binomial with a varietal form; common name of the disease, Corynose twig blight.

8. No ascogenous stage was observed in the cultures.

9. To control the disease, the dead and diseased twigs should be so pruned that the cuts remove any perennial mycelium residing in them. All pruned rubbish should be burned at once, since the conidia may remain viable within incised twigs for 7 months.

MASSACHUSETTS STATE COLLEGE,
AMHERST, MASS.

WATER BLISTERING OF WOUND DRESSINGS

RUSH P. MARSHALL

The painted surface frequently blisters when large, freshly made cuts on trees are painted thickly with tacky, impervious materials, such as heavy tar or asphalt (Fig. 1). This is particularly true when the material used does not adhere well to wet wood. While wound dressing may blister from other causes, the trouble appears for the most part to be produced by moisture, which collects between the wood surface and the protective skin of paint in the form of water blisters of variable size. In time these blisters tend to break and, so, furnish points of entrance through which insects and fungi may attack the wood.

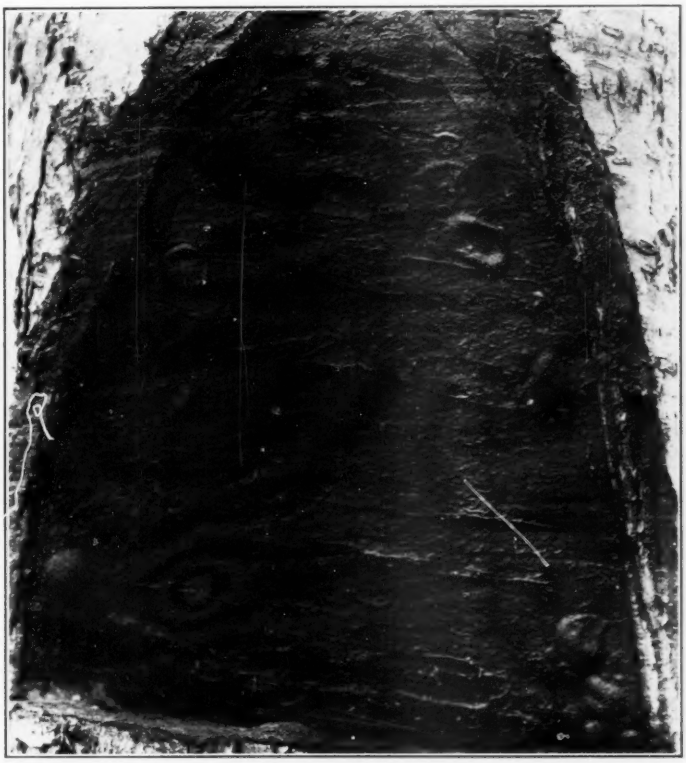

FIG. 1. Blistering of a heavy asphalt wound dressing.

In determining a method of observing something of what takes place when a cut is covered by an impervious coating, the writer is indebted to

F. A. Bartlett, who conceived a plan of covering wounds with glass. In executing this plan, both large and small glass-covered wounds were made and observed, not with the idea that they would duplicate or parallel conditions obtaining beneath the painted surface but rather in the hope that the results obtained might be indicative of what took place beneath the paint. The experiment here described was conducted at Stamford, Conn., in an open stand of mixed hardwood. The upper part of the site used is well drained but springy; while, in the lower reaches, where the birch and maple are located, the ground is wet during the greater part of the year. The trees selected for the larger cavities had a breast height diameter of 12 to 14 in., and, for the smaller cavities, a diameter of 6 to 8 in. The following tree species were included: *Acer rubrum*[1] L., *Hicoria alba* (L.) Britton, *H. ovata* (Mill.) Britton, *Betula lenta* L., *B. lutea* Michx. f., *Fagus grandifolia* Ehrh., *Liriodendron tulipifera* L., *Nyssa sylvatica* Marsh, *Quercus alba* L., *Tilia glabra* Vent., and *Ulmus americana* L.

The larger glass-covered wounds were all made very skillfully by L. Strout. The first 4 of these were put in during April, 1927, and 7 additional ones added during 1928 and 1929, making a total of 11 of the larger cavities. These wounds were made in sound wood at breast height on the trunk. They were 12 to 14 in. long, 6 to 8 in. wide, and were dug 4 to 6 in. into the wood. They were shaped as are cavities ordinarily employed by tree surgeons. In executing the work, the window glass used to cover the opening was first cut to form a lenticular pane. This was held against the trunk with its axis parallel to the axis of the tree and its outline traced on the bark with chalk. The area of wood within this line was then chamfered out to form a flat face so that the glass could be set into the opening to about $\frac{1}{4}$ in. below the cambium line. Next, leaving a $\frac{1}{2}$-in. shoulder to support the pane, the center of the area was chiseled out to form a regulation cavity. The outer edge of the cut was sealed with surgeons' tape and then covered with fitted strips cut from asphalt roofing paper. The edge and shoulder were then faced deeply with asphalt putty and the pane put into place. In this way the glass was set into a more or less flexible matrix, so as to allow slight play during the stress and strain of the trunk, occasioned by wind. The cavity was, however, made impervious to water and air. It was thus possible to look through the glass window and observe what took place behind an impervious covering (Fig. 2, A).

The callus healed well over the edge of the glass and its growth sealed the wound more and more tightly. As the callus grew it clamped down about the glass, leaving no room for play, and the pane ultimately snapped with the torsion of the trunk in the high wind or from the pressure of tree

[1] Nomenclature from: Sudworth, G. B. Check list of the forest trees of the United States, their names and ranges. U. S. Dept. Agr. Misc. Circ. 92. 1927.

growth (Fig. 2, B). In making later cavities heavy glass from automobile wind shields was substituted for window glass, but it failed to show advantage and in spite of its greater thickness, generally broke at the end of about 2 years.

FIG. 2, A. One of the large glass-covered wounds photographed shortly after its completion. The tree is yellow poplar. B. The same wound 2 years after its completion. The callus has clamped down on the glass, and the pane has broken.

In order to permit similar observation on numerous trees without the expense of setting large glasses, or inflicting so much damage to valuable trees, an attempt was made to use small wounds covered with regulation laboratory watch glasses. These could be set within a few minutes at little cost and with a minimum of injury to the trees. They had the further advantage of permitting trees as small as 6 or 8 in. D. B. H. to be used. The modified wound was bored with a bit. It was started with an extension bit, set to cut a 2-in. hole. This was drilled deeply enough to remove

about ¼ in. of sapwood. Boring was then continued almost entirely through the trunk, using the 1½-in. auger. The hole was then centered to the far side of the trunk with a ⅜-in. twist drill. When the point of the small drill came through on the far side of the tree, the extension bit was again used to bore out another 2-in. hole ¼ in. deep, this time on the far side of the trunk. In this way a 1½-in. auger hole passed entirely through the trunk and was faced with a shallow 2-in. hole on the other side. Two-inch watch glasses were set into these 2-in. openings with either asphalt putty or grafting wax (Fig. 3, A).

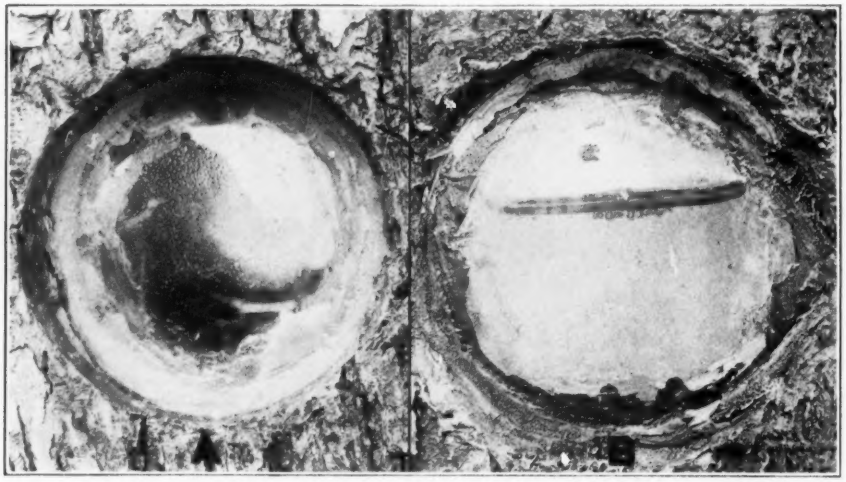

FIG. 3, A. One of the smaller glass-covered wounds photographed shortly after completion. Note the sweating of the inner surface of the glass. The wound is in white oak. B. With the rise of sap in the spring some of the smaller wounds become partially or entirely filled with liquid. The wound is in yellow birch.

Although they were easily installed, and their watch-glass windows withstood breakage well in most cases, the smaller wounds were subject to three faults, which made them far less satisfactory than were the larger cavities: (1) The material used in sealing the glasses into the openings frequently loosened and permitted the cavities to leak. (2) The cavities were too small for accurate results. (3) Close observation of what took place within them was difficult. This latter fault was evident at the outset of the experiment when in several early trials the holes were bored only part way through the trunk and the opening closed with a single watch glass. The small size of the opening and the sweating of the inner surface of the glass hindered observation. Cavities covered by a single watch glass were abandoned in favor of the method already described, because the use of two glasses admitted light, both front and rear, and facilitated inspection, particularly

when a flashlight was used to illuminate the opening from one side while observation was made from the other. Even then, the smaller cavities did not show clearly what took place at the back of the wood, since, at best, one could see only the sides of the auger hole. Despite their faults, 16 of the smaller cavities were made in 1927 and an additional 25 in 1928.

Observation of both the large and small cavities showed that moisture tended to collect behind the glass coverings. This appeared to have come from two sources, namely: (1) Sweating, due to condensation of moisture from within on the inner surface of the glass (Fig. 3, A), and (2) actual liquid that flowed from the wound. With the rise of the sap in the spring the maple and birch showed more or less profuse bleeding, while other species showed but little. In the case of the maple and birch, the watch-glass cavities tended to become partially or entirely filled with sap in the first spring following their making (Fig. 3, B). This was particularly true for both the black and the yellow birch, which bled so profusely that the watch glasses were often forced out by the pressure under which the sap flowed. In the case of the large wounds the cavities were not observed to have become more than $\frac{1}{3}$ filled with liquid. Bleeding of the red maple occurred in the middle of March, while in the black and yellow birch it did not take place until the latter part of April. In either case it was

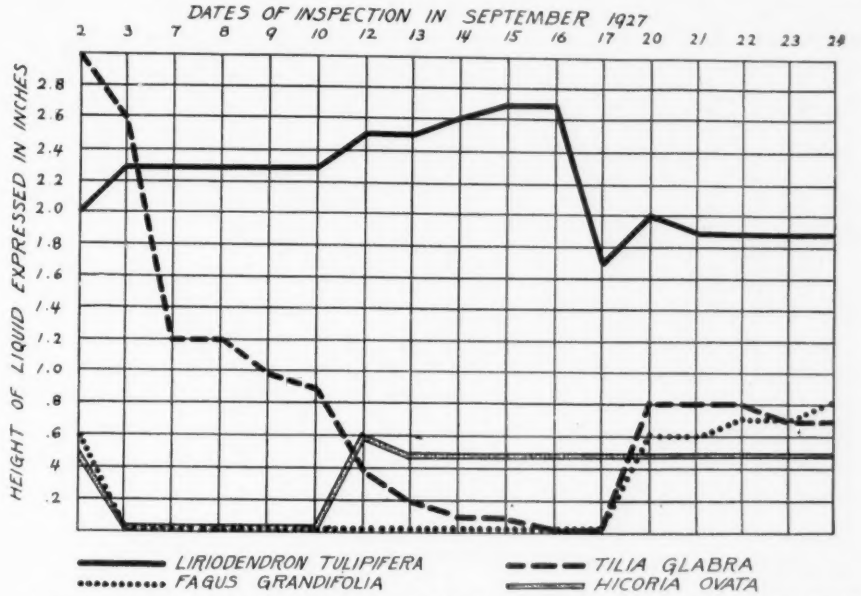

FIG. 4. Graph showing an example of the daily variation of the height of liquid which collected in the 4 original cavities made in the spring of 1927. Note that the curves lack correlation.

of relatively short duration, and within a few weeks the sap was taken up again by the wood and more or less disappeared from the cavities.

Throughout the year an unexpected accumulation of liquid was frequently evident in the larger wounds. It was so much less in volume than the spring flow that it was seldom noticeable in the smaller wounds. The volume of the liquid was found to vary from day to day (Figs. 4 and 5). Although a number of observations were made during 3 years, the writer was unable to account for the occurrence of this flow or to correlate it with either external or internal phenomena. The time it took place in some of the trees was not necessarily simultaneous with that of its appearance in others, nor did it seem to be closely connected with atmospheric conditions or with soil moisture. It took place in both the growing and dormant seasons and in both wet and dry weather.

Fungus growth occurred within several of the cavities. It was not investigated by the removal of the glasses. Observation through the glass seemed to indicate that, among other forms, Mucor, Penicillium, Graphium, Ceratostomella, and a pink form of yeast were present. Basidiomycetes were not observed.

Aside from the universally recognized function of protection against invasion of insects and fungi, no other reason is perhaps more generally given for the application of dressings to tree wounds than that of keeping out water. The well-known slogan of the painters, "Save the surface and you save all", appears to have been carried over into at least the subconscious mind of those who apply paint to trees as well as those who make use of it on houses. The scientist as well as the gardener has frequently been remiss in this regard, as is shown by the fact that articles dealing with the subject of wound dressings sometimes stress the importance of paint in keeping moisture out of the wound.

The water that collected behind the glass in these experimental cavities was much greater in amount than that occurring within the water blisters common to thick, impervious dressings. This would lead one to question the statement that one of the purposes of a wound dressing is to keep out moisture. We all know that moisture is ever present within the wood of the living tree. More than that, it is possible for active bleeding to take place at any season of the year. This bleeding is not necessarily restricted to the sapwood.

The present experiment leads us to suspect that, theoretically, the ideal wound dressings for heartwood will be a material, not too perfectly impervious, but rather one that will permit some passage of water from the wound through the dressings, thereby tending to prevent its collection be-

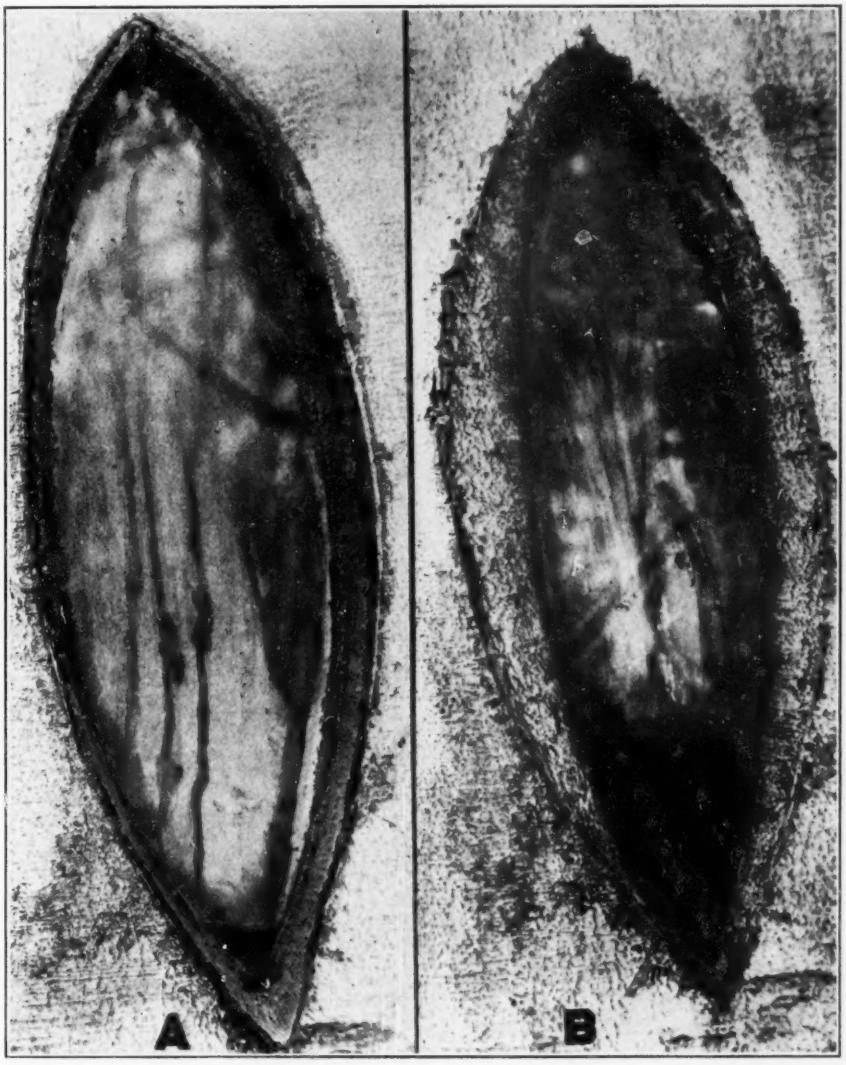

FIG. 5, A. A cavity in a beech made in the spring of 1928 and photographed shortly after completion. Note the flow from the heartwood and the collection of one inch of liquid. B. The same cavity photographed in the fall of 1929. Three and one-fourth inches of liquid were present in the cavity.

neath the surface with resultant blistering. It seems to indicate that in using any of the many excellent tree paints, which are highly impervious, we should guard against applying the material in an unreasonably thick coating.

DIVISION OF FOREST PATHOLOGY,
BUREAU OF PLANT INDUSTRY, AND
OSBORN BOTANICAL LABORATORY,
YALE UNIVERSITY, NEW HAVEN, CONN.

AGGLUTINATION STUDIES ON PHYTOMONAS MALVACEARA

O. B. WILLIAMS AND H. B. GLASS[1]

The present work was undertaken in the endeavor to determine whether or not there are distinct serological strains of the plant pathogen, *Phytomonas malvaceara* (E.F.Sm.) Com. S.A.B., the causal organism of angular leaf spot of cotton. A considerable amount of serological work has been done on plant pathogenic bacteria, chiefly by St. John-Brooks, Nain, and Rhodes (11), Sharp (10), Link and Sharp (7), Link and Hull (5), Link and Link (6), Link and Taliaferro (8), and Goldsworthy (1), as a result of which it has been shown that serological methods are of value in recognizing and in separating the various species. The possibility of the existence of serological strains within a single species, however, has not been the subject of any extensive amount of work.

Fourteen strains of *Phytomonas malvaceara*, isolated from field cotton plants by Dr. I. M. Lewis, in 1928, and tested for pathogenicity by him, were used for the study. These were grown at room temperature on nutrient sucrose agar, a medium that has been found especially suitable for the cultivation of this organism. It was early found that killed organisms could not be successfully used for the production of a high-titre serum. After a number of experiments with varying dosages and varying intervals of time between doses, it was found that an immune serum with a satisfactory titre could be produced in rabbits by the intravenous inoculation of living organisms. The method finally followed consisted of washing down, just before inoculation, the growth from a heavily seeded sucrose agar slant with physiological sodium chloride solution and then diluting to a turbidity approximately equivalent to 1,000 million organisms per ml. Three doses of 0.5, 1.0, and 1.5 ml. of this suspension were given at 10-day intervals, followed by a series of 3 doses of 1.0 ml. each at 3-day intervals, and bleeding 5 days after the last inoculation. The pooled serum of 2 rabbits immunized by this method had a titre of 1:6,000 for the homologous strain (No. 45). This serum was preserved by the addition of an equal volume of glycerin.

Agglutination tests with the various strains as antigens were carried out in dilutions of this serum ranging from 1:300 to 1:18,000, according to the usual methods. The tests were incubated at 45° C. for 2 hours and held over night in the ice box before recording the results. All tests have been

[1] Since this report was submitted for publication there has appeared a paper by Horgan (Jour. Bact. 22: 287–293. 1931) in which it is shown by agglutinin-absorption tests that African strains of this organism also constitute a serologically homogeneous group.

repeated at least 3 times. Considerable difficulty was encountered in the early experiments, due to spontaneous agglutination. This was finally controlled by the use of 0.45 per cent salt solution instead of the customary 0.86 per cent. A single strain of *Phytomonas bowlesii* Lewis & Wats. was included in the series, with consistently negative results. The results obtained are shown in table 1.

TABLE 1.—*Results of agglutination tests*

Strain	Serum dilution									
	300	600	1,200	1,500	3,000	6,000	9,000	12,000	18,000	Control
44	++++	++++	++++	++++	+++	++	+	+	+	–
45	++++	++++	++++	+++	++	+	–	–	–	–
46	++++	++++	+++	+++	++	++	+	+	–	–
47	++++	++++	+++	++	+	+	–	–	–	–
48	++++	++++	++++	+++	++	+	+	–	–	–
49	++++	++++	++++	+++	++	+	+	–	–	–
50	++++	++++	+++	++	+	+	+	–	–	–
51	+++	+++	++	+	+	–	–	–	–	–
52	+++	++	++	+	+	–	–	–	–	–
53	++++	++++	+++	++	+	+	–	–	–	–
54	++++	++++	++++	++	++	+	+	–	–	–
55	+++	++	+	–	–	–	–	–	–	–
56	++++	++++	+++	+++	++	+	+	–	–	–
57	++++	+++	+	+	–	–	–	–	–	–
Phy. bowlesii	–	–	–	–	–	–	–	–	–	–

++++ = Complete agglutination.

The variations in titre obtained suggested the possibility that serological strains were present, hence absorption tests were carried out. In making the absorptions with the nonhomologous strains the suspensions were added, and, after absorption and centrifuging, reabsorption was done until all the agglutinins were removed for the strain used. This required from three to four repetitions of the process. Two strains, besides No. 45, the homologous strain, were used for these absorptions, i.e., No. 44, which the cross agglutinations had shown to be of the highest titre, and No. 55, similarly shown to be of the lowest titre. Cross agglutinations of all the strains were made with these three absorbed sera.

Although it would appear from the results given above that the organisms might be separated into at least two groups, one of high and the other of low agglutinating ability, sera completely absorbed for each of the three strains mentioned were also absorbed for each of the remaining strains, with a single exception. Strain No. 44 was agglutinated in a dilution of 1:600 of serum absorbed with strain No. 45. Since it was observed, especially with sera absorbed with strain No. 55, that, when a serum that had been

incompletely absorbed was used, differences in titre similar to those given in the tabulation were obtained, it is felt that the exception mentioned was due to incomplete absorption and is not, therefore, significant.

DISCUSSION

Tittsler and Lisse (12) have shown that agglutination in strains of *Salmonella pullorum* (Rettg.) Bergey *et. al.* is correlated with the rate of electrophoretic migration of the organisms in suspension, the higher being the rate of migration, the lower the titre obtained. This would seem to mean that in the present work the differences of titre in cross agglutinations depend upon differences in the charges on the organisms, since absorptions reveal no constant differences between them. The results here reported show, as Krumwiede, Cooper, and Provost (3) maintain, that no valid conclusions can be based on cross agglutinations alone, but that absorption tests also are necessary. Whereas earlier workers, like Hooker (2), found absorption tests confirmed cross-agglutination results, this has been gravely questioned by later workers. Differences in agglutinability may be present within strains of a single species, which are neither clear nor definite enough to provide criteria for separation of the strains into groups, as has been pointed out by Robinson (9). The work of Tittsler and Lisse (12) affords at least one explanation of these differences.

Lewis (4) has shown that strains of *Phytomonas malvaceara* are culturally quite uniform. The report here presented indicates that they are also serologically uniform.

All of the strains employed in this work appeared to be of the smooth type. The possibility of serological differences due to dissociation is being further investigated in this department.

SUMMARY

It was found possible to produce a high-titred agglutinating serum for *Phytomonas malvaceara*.

Cross reactions with heterologous strains of the same organism showed considerable differences in agglutinating titre but absorption tests failed to establish any fundamental serological differences between the strains used.

DEPARTMENT OF BACTERIOLOGY,
THE UNIVERSITY OF TEXAS,
AUSTIN, TEXAS.

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AN ANTHRACNOSE OF THE JUJUBE[1]

J. J. Taubenhaus and Walter N. Ezekiel

The jujube, *Zizyphus jujuba* Mill., has been grown in Texas more than 50 years, chiefly as an ornamental.[2] The recent introduction of improved varieties, however, has encouraged planting the jujube for its fruit. Jujubes are adapted to a variety of soils, and a crop is set, even in dry years.

In August, 1926, a heavy premature shedding of the fruit was noticed in a jujube orchard near College Station, Tex. (Fig. 1, A). It was at first thought that the shedding might have been caused by the dry weather. However, some of the fruits on the ground were examined and found to be studded with numerous small, dark superficial spots (Fig. 1, D). Immature as well as ripe fruits, still clinging to the trees, were similarly affected but the foliage and limbs appeared sound. The spots first appeared as small, round, dark dots; developing into smooth or slightly sunken circular spots from 4 to 6 mm. in diameter (Fig. 1, B and C). In cases of heavy infections they frequently coalesced. The fruits on the ground were found to be more generally spotted than those still clinging to the trees. Sectioning such fruit, it was found that the interior tissues had darkened (Fig. 1, E) to a deep olivaceous color and were permeated with fungus hyphae. Severely diseased fruit shrivelled and finally became mummified (Fig. 1, F).

LOSS

To determine the approximate loss from the disease, a count was made early in September of the infected fruits on 3 trees selected as representative. It was found that 40 per cent of the fruits were diseased and had fallen to the ground; 30 per cent were variously spotted but clung to the trees; while the remaining 30 per cent were apparently sound. The disease was rather scarce during 1927 and 1928, was again destructive in 1929, and somewhat less serious in 1930.

CAUSE

Usually, no spores could be found on the surface of affected fruits; but when infected fruits were held in moist chambers over night they became covered with salmon-color layers of typical Gloeosporium spores. Pure cultures of a Gloeosporium were obtained from the spores and from bits of the interior tissues of diseased fruits (Fig. 1, H). Spores from these cultures were used in the inoculations reported below.

[1] Published with the approval of the Director as Contribution No. 155, Technical Series, of the Texas Agricultural Experiment Station.

[2] Lanham, W. B. Jujubes in Texas. Texas Agr. Exp. Sta. Circ. 41: 1926.

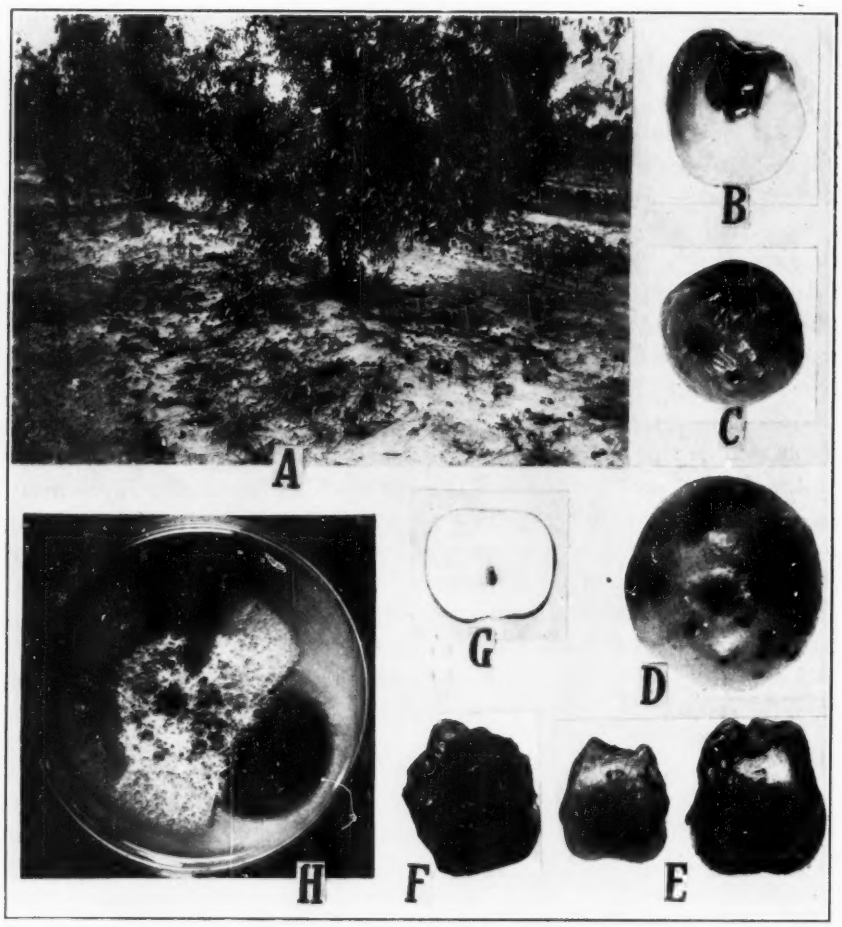

FIG. 1. Jujube anthracnose. A. Premature shedding of jujube fruit due to anthracnose. B, C, and D. Anthracnose on jujube fruits, showing variation in size of spots. E. Cross-section through infected jujube. F. Mummied, infected jujube. G. Cross-section through normal jujube. H. Gloeosporium growing from infected jujube tissue (Aspergillus contamination to the right).

On September 2, apparently sound jujube fruits on the trees were inoculated. Branches with healthy fruit were enclosed in large paper bags, sprayed with a suspension of spores, and the ends of the bags then tightly closed to prevent drying. Five hundred jujube fruits were inoculated in this way. By September 12, 86 per cent, or 432 fruits, showed definite signs of infection with symptoms resembling those found on naturally infected fruit, while only 7 per cent of 390 uninoculated fruits had developed anthracnose spots.

TABLE 1.—*Inoculations of various fruits and roots in moist chambers in the laboratory, using spores of the jujube Gloeosporium from pure cultures*

Fruits and roots inoculated	Date of inoculation	Date of observations	Results with inoculated fruits and roots			Results with check, uninoculated fruits and roots		
			Total number	Number infected	Percentage	Total number	Number infected	Percentage
	1926	*1926*						
Jujubes	Sept. 3	Sept. 15	250	108	43	250	7	3
Apples	Oct. 12	Oct. 25	25	25	100	25	0	0
Figs	Nov. 16	Nov. 22	25	21	84	25	0	0
Grapes	Oct. 29	" 4	6 bunches	6 bunches	100	6 bunches	0	0
Japanese persimmons	Nov. 16	Dec. 6	25	25	100	25	0	0
Grapefruits	Oct. 21	Oct. 29	12	12	100	12	0	0
Oranges	" 26	Nov. 3	12	12	100	12	0	0
Tomatoes, green	" 15	Oct. 19	25	25	100	.....	.....	.....
" pink	"	"	25	25	100	25	0	0
Sweet peppers	" 29	Nov. 15	24	24	100	24	0	0
Squashes	Nov. 16	" 29	12	0	0	12	0	0
Radishes	"	"	24	0	0	24	0	0
Turnips	"	"	24	0	0	24	0	0

Further inoculations in the laboratory are summarized in table 1. Sound jujube fruits were disinfected for ½ minute in a solution of 1:2,000 mercuric chloride in 25 per cent alcohol and then rinsed several times in sterilized water. Some of these fruits were then inoculated through needle punctures in the epidermis with spores from a pure culture of the jujube Gloeosporium. Similar inoculations were made on 11 other possible hosts.

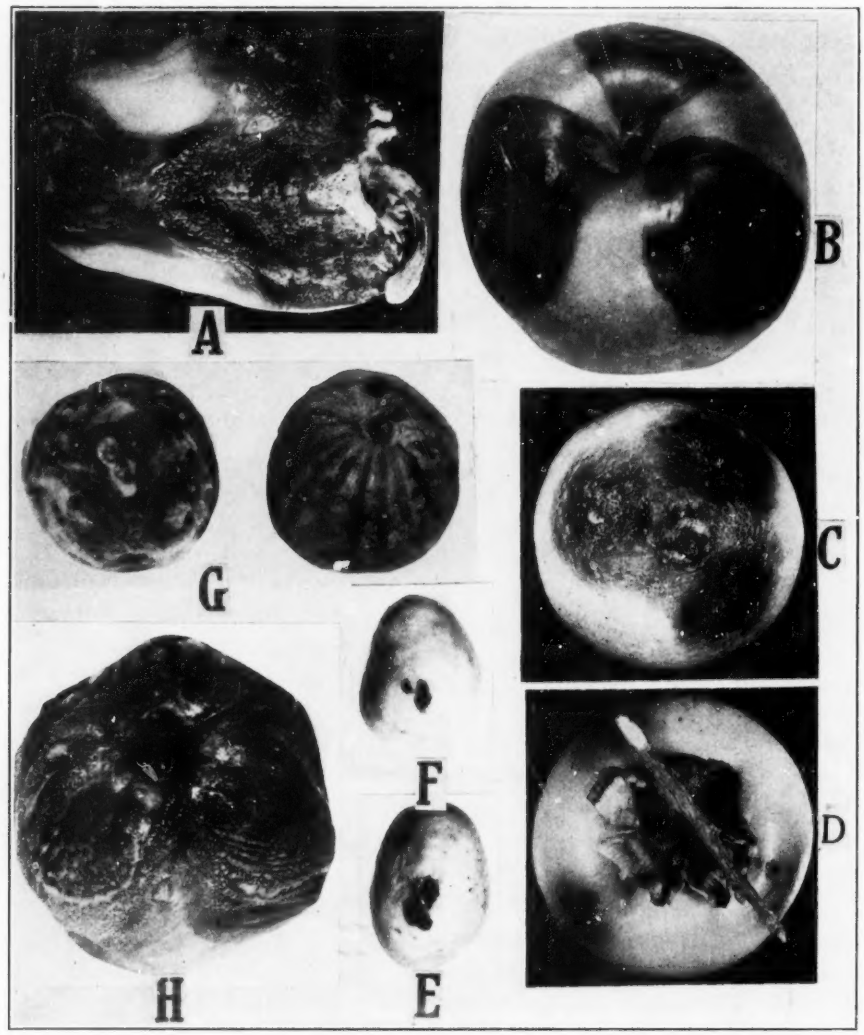

FIG. 2. Artificial inoculation of various fruits with Gloeosporium isolated from jujubes. A, on pepper; B, on apple; C, on orange; D, on Japanese persimmon; E and F, on jujube fruits; G, on fig; H, on tomato.

As listed in table 1, typical anthracnose infection was secured on the jujube fruit (Fig. 2, E and F) and on apples (Fig. 2, B), figs (Fig. 2, G), grapes, Japanese persimmons (Fig. 2, D), sweet peppers (Fig. 2, A), tomatoes (Fig. 2, H), oranges (Fig. 2, C) and grapefruit. No infection was secured with radish, turnips, or squash. From these and the previous inoculations, it appears that the Gloeosporium isolated from infected jujube fruit in the orchard is responsible, at least in part, for the jujube disease under consideration.

IDENTITY OF THE FUNGUS

On oatmeal agar, the jujube Gloeosporium formed salmon-color masses of typical unicellular spores, but no setae; while setae were numerous on jujube agar (50 gm. jujube fruits to 1,000 cc. tap water, steamed and filtered, plus 30 gm. agar). The fungus did not develop setae on artificially inoculated jujubes, figs, and apples, while they were numerous on inoculated sweet peppers and tomatoes. The fungus was grown on other media also, but no perfect stage was found there nor on overwintered mummied fruits that were examined. From the inoculations reported in table 1, however, and from the behavior of the fungus in culture, the jujube Gloeosporium probably is the same as or closely related to the Gloeosporium stage of *Glomerella cingulata* (Stoneman) S. and v. S., the cause of the bitter rot of apples, which has been shown[3,4] to attack many of the fruits that were found susceptible to this jujube fungus.

Recent isolations from diseased jujube fruit have yielded several fungi, including a species of Diplodia, in addition to the Gloeosporium mentioned, and further studies with these other organisms are in progress.

SUMMARY

A serious anthracnose disease of jujube fruit, resulting in premature shedding, was found to be caused, at least in part, by a Gloeosporium. Normal jujube fruits, as well as apples, figs, grapes, Japanese persimmons, peppers, tomatoes, grapefruits, and oranges were successfully inoculated with spores from pure cultures isolated originally from jujube fruit, and the organism was recovered from the inoculated fruit. No asci have been found on infected jujube fruit or in culture, but in general appearance and behavior the fungus appears to be the same as or closely related to *Glomerella cingulata*, the cause of bitter rot of apples.

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A DISEASE OF YOUNG COTTON PLANTS CAUSED BY SCLEROTIUM ROLFSII[1]

WALTER N. EZEKIEL AND J. J. TAUBENHAUS

Sclerotium rolfsii Sacc. is wide-spread in the South and damages a large number of hosts. This fungus has been reported on cotton plants,[2,3] which, however, have generally been considered resistant. The fungus has also been recorded as causing boll rot of cotton in Florida.[4] The present paper reports a definite disease of young cotton plants, proved to be due to *S. rolfsii*.

On July 1, 1929, cotton was planted in experimental plats at College Station, Tex., in a field of Lufkin fine sandy loam. Following unusually wet weather early that summer, some of the seedlings were found dying apparently from the attack of *Sclerotium rolfsii*, and by July 28 many plants were dead. Before the plants were thinned, the fungus spread along the rows often for distances of more than 1 foot, in one instance killing 18 plants in a space of 9 inches along the row. This severe attack, limited to restricted areas in the rows, was characteristic of the disease. Slow spread continued until about August 20, when it was checked possibly by the prolonged dry weather, although the plats were watered frequently.

Individual infected plants were readily recognized (Fig. 1, a to d). The fungus attacked and girdled the base of the stem; this was followed by rapid wilting and death of the plant. The infected part of the stem appeared water-soaked at first and then became constricted at about the ground level. The cortex occasionally split along the constricted area. Even after the upper part of the plant wilted and died, the roots remained unaffected and apparently normal.

The fungus traveled from plant to plant superficially on or through the soil. The mycelium about the base of freshly infected plants was white, usually mat-like, and readily visible to the naked eye, and sclerotia characteristic of *Sclerotium rolfsii* were produced usually by the time the plant died. The sclerotia developed superficially on the base of affected plants and on the soil (Fig. 1, a); they were spheric in shape and at first white, becoming yellow and then brown with age. Some plants did not show the

[1] Published with the approval of the Director as Contribution No. 156, Technical Series, of the Texas Agricultural Experiment Station.

[2] Rolfs, P. H. Report of the horticulturist. Fla. Agr. Exp. Sta. Ann. Rpt. **1896**: 21–52. 1897.

[3] Taubenhaus, J. J. Recent studies on *Sclerotium rolfsii* Sacc. Jour. Agr. Res. **18**: 127–138. 1919.

[4] Walker, M. N. Cotton diseases in Florida. Fla. Agr. Exp. Sta. Bul. 214. 1930.

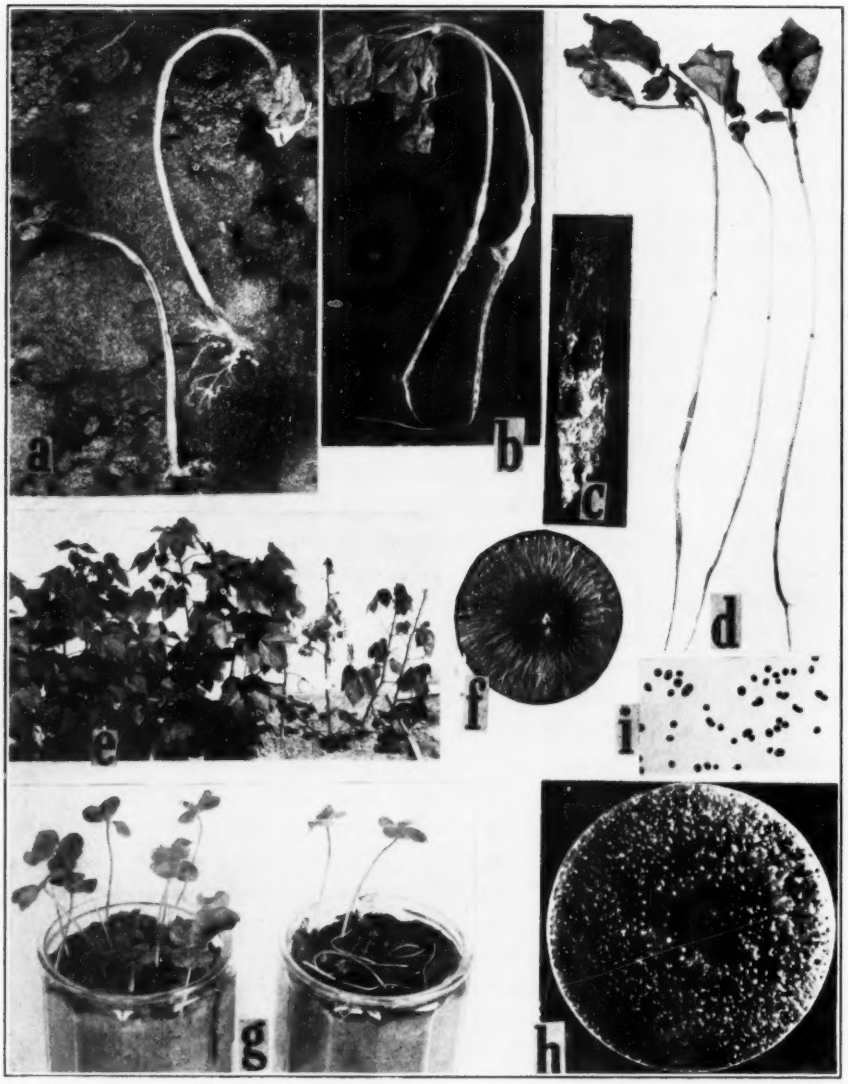

FIG. 1. *Sclerotium rolfsii* stem rot of cotton seedlings: a, two seedlings artificially inoculated with *S. rolfsii;* b and d, naturally infected seedlings; c, enlarged view of one of the plants from b; e, a mature cotton plant dying from natural infection by *S. rolfsii;* f, young culture of *S. rolfsii* from a single sclerotium from i; g, right, seedlings artificially inoculated with *S. rolfsii,* and, left, uninoculated plants; h, pure culture of *S. rolfsii* isolated from an infected cotton seedling; i, mature sclerotia from h.

strands on the surface of the stems in the field but produced a copious growth of mycelium and sclerotia when placed in moist chambers.

INOCULATION

Cultures made from surface-sterilized tissue from diseased stems, as well as cultures from the sclerotia, yielded growth typical of *Sclerotium rolfsii* (Fig. 1, h) and were similar to cultures of this fungus isolated previously from infected carrots.

Strains of *Sclerotium rolfsii* from cotton and carrots were grown on sterilized cotton stems and used in inoculations of cotton plants in the laboratory. Twelve containers filled with sterilized soil were planted each with 12 cotton seeds previously delinted with sulphuric acid and surface-sterilized in mercuric chloride solution. After the plants were up, plants in 4 containers were inoculated with the strain of *Sclerotium rolfsii* from infected cotton plants, and plants in 4 other containers with the strain from carrots. The containers were covered overnight with bell jars. Within 3 to 5 days after inoculation, the inoculated plants wilted and died, while the check plants in the 4 other containers remained healthy (Fig. 1, g). The symptoms produced on these inoculated plants were similar to those observed in the field, and the fungus was readily reisolated from the diseased seedlings. A second series of inoculations, of somewhat older cotton seedlings, was equally successful and proved that *Sclerotium rolfsii*, whether isolated from cotton or from carrots, may cause a stem rot of young cotton plants.

In 1928, several attempts had been made to inoculate mature cotton plants in another field, in connection with other experiments. These cotton plants were fully grown and had already produced well-developed bolls. The *Sclerotium rolfsii* strains used were isolated originally from carrot and from guar, *Cyamopsis tetragonoloba* L. Fresh cultures on sterilized cotton stems were placed next to the base of the plants, covered with moist straw, and water applied at intervals. No infection resulted. It is to be noted, however, that a mature cotton plant found wilting in an experimental plat in 1930 had apparently been killed by *S. rolfsii*, the only organism isolated from it in cultures (Fig. 1, e). These experiments suggest that *S. rolfsii* probably attacks older cotton plants only rarely, although it may destroy young seedlings or plants ranging from a week to 10 weeks old.

EFFECT OF SOIL DISINFECTANTS

The experimental plats in which the disease was found were being used in tests of the efficiency of various soil disinfectants for the control of Phymatotrichum root rot. On August 19, 440 plants dead or dying from

the attack of *Sclerotium rolfsii* were counted in the 30 plats of this field. There were some affected plants in most of the plats; however, there appeared to be significantly fewer diseased plants in the plats in which various organic-mercury compounds had been incorporated into the surface soil than in adjoining check plats. This was the case with K–1–X, PMA, Semesan, No. 664, and Bayer Dust. There were 275 diseased plants in 7 check plats, each about 27 feet long and 20 feet wide, while only 63 affected plants were found in the 14 treated plats, each 20 feet long and 20 feet wide. More plants with stem rot were found in each check plat than in both the adjoining treated plats. On the other hand, in the 6 plats treated with other materials, such as copper sulphate, copper carbonate, and iron sulphate, there was no consistent decrease of infection in the treated plats, which in some cases had more diseased plants than in the corresponding checks. Stucky[5] has previously reported promising results obtained by Higgins with Du Pont K–1–P applied about the base of pepper plants attacked by *S. rolfsii*.

SUMMARY

A stem rot of young cotton plants was found to be due to the fungus *Sclerotium rolfsii*. This was proved by artificial inoculations of young seedlings grown in sterilized soil in the laboratory. Artificially inoculated mature plants, in the field, however, did not become infected. Fewer diseased plants occurred in field plats in which organic-mercury compounds had been applied than in the check plats.

[5] Stucky, H. P. Director's report. Ga. Agr. Exp. Sta. Ann. Rpt. 41: 24. 1928.

A SCLEROTINIA LIMB BLIGHT OF FIGS[1]

J. J. Taubenhaus and Walter N. Ezekiel

During September, 1926, a serious limb blight was found in many orchards in Galveston County, Tex., where the Magnolia fig is grown extensively for canning purposes. Growers claimed that the trouble was the result of the previous severe winter. However, the affected limbs died too late in the season for winter injury; there were no deep cracks in the bark of the limbs or main trunks, which characterize frost damage; and several growers stated that they had seen a similar blight in previous years, in seasons following mild winters.

The disease became noticeable as a sudden wilting of the foliage, followed by dying of the affected branches (Fig. 1, A). Other limbs were gradually involved until large portions of the trees were affected. The trunks of some trees also were attacked. More rarely, the base of a tree was infected and girdled. Examination of the diseased areas showed that they were of a water-soaked appearance and overrun by a thick, white fungus growth. The exterior and interior of the affected limbs soon became studded with numerous sclerotia (Fig. 1, B to E), which were found to resemble sclerotia of *Sclerotinia sclerotiorum* (Lib.) Mass.

Search was made in several orchards to locate the source of infection. In the vicinity of infected trees, partly buried sclerotia were found bearing mature apothecia, which were discharging clouds of ascospores. The apothecia, asci, and ascospores closely resembled those of *Sclerotinia sclerotiorum*. A culture from the ascospores was used successfully in the inoculation experiment referred to below. It therefore seems probable that ascospores constituted inoculum for natural infection.

Pure cultures were secured from bits of tissue of affected fig limbs, from sclerotia found on diseased fig limbs, and from ascospores. Cultures of *Sclerotinia sclerotiorum* were available also from celery, beans, lettuce, and snapdragons. These 7 strains of the fungus were grown on sterilized bean pods, and were used in October, 1927, in the inoculation of normal, 2-year-old potted fig plants growing in 12-inch pots. These plants were from rooted cuttings secured from the nursery of the college campus and had not previously been exposed to the fungus. The inoculum was placed close to the foot of the plants and covered with a loose layer of moist oat straw. Seventy potted fig plants were inoculated, using 10 plants for each strain of the fungus, and 10 uninoculated plants were left as checks. High percentages of infection resulted from all the strains of Sclerotinia

[1] Published with the approval of the Director as Contribution No. 154, Technical Series, of the Texas Agricultural Experiment Station.

FIG. 1. Sclerotinia limb blight of figs. A. Fig tree killed by *Sclerotinia sclerotiorum*. B, C, D, and E. Blighted fig limbs studded with sclerotia of *S. sclerotiorum*.

used, while not a single infection occurred in the check plants. The appearance of the artificially inoculated plants closely resembled that of naturally-infected limbs in the orchards. The Sclerotinia was readily recovered from the inoculated plants.

In the field blighted fig limbs, particularly the older ones, were frequently overrun by *Tubercularia fici* Edgerton. In advanced cases, evidence of the presence of the Sclerotinia was sometimes obscured, making it appear that the Tubercularia had caused the principal damage. Condit and Stevens[2] described a "die-back" of the fig in California, which they attributed to *Botrytis cinerea* and *Sclerotinia libertiana*. We found no injury from Botrytis on Texas figs, nor did the limb blight described here resemble the condition usually spoken of as die-back. *Sclerotinia sclerotiorum* appeared to be the chief, if not the only, cause of the limb blight.

Search was made to determine whether other host plants growing near affected trees in the orchards were also attacked by the fungus. Plants of *Amaranthus retroflexus*, a common weed in many of the orchards, were found heavily infected and in different stages of wilting. In other orchards, bean and lettuce plants growing in the vicinity of infected trees were also attacked by Sclerotinia. Cultures were made from infected tissues of these hosts, and the resultant growth was similar to *Sclerotinia sclerotiorum* isolated from infected fig limbs.

SUMMARY

A blight of fig limbs is described. The disease was shown to be due to *Sclerotinia sclerotiorum*. Natural infection in the field was apparently from ascospores discharged by apothecia found near infected trees.

[2] Condit, I. J., and H. J. Stevens. "Die-back" of the fig in California. Month. Bul. State Comm. Hort. Calif. 8: 61–63. 1919.

PHYTOPATHOLOGICAL NOTES

Data to be Noted in Studying Heart Rots of Living Trees.—In a rather extensive study of heart rots through a period of years the writer has found the following points of great value in studying such rots. These items are given in the hope that they may be of interest to other workers in this and related fields of research. It is not claimed that these data represent all that should be recorded, but they will give a good idea of the heart rot and what to expect under various conditions of growth, site, timber, etc.

General field observations

(1) Soil, slope, moisture, altitude, etc., where infected trees are found.
(2) Age and number of trees and species attacked by each heart rot.
(3) Diameter and height of rot in tree.
(4) Methods of entrance to tree, through roots, scars at butt, broken limbs, wormholes, etc.
(5) Position of sporophores (a) on ground, (b) on roots, (c) on trunk, (d) on branches, etc.
(6) External evidence of heart rot (a) punks, (b) hollows, (c) swelled butts, etc.
(7) Effect on growth of leaves, twigs, trunk, and roots.
(8) Odor, if any, of rot when freshly cut.
(9) Condition of rot in stump on weathering: fibrous, cubical, agglutinated, etc.
(10) In the case of conifers, infiltration of pitch, if any, in front of advancing fungus (a) in trunk, (b) in infected branches.
(11) Appearance of top of stump when freshly cut as firm, sieve-like, hollows, etc.
(12) Character of rot: wet, dry, spongy, brittle (a) when fresh, (b) when dry.
(13) Extension, if any, of heart rot into sapwood.
(14) Continuation of growth of rot in felled tree.
(15) Is the rot a root rot, a butt rot, or a top rot, or does it occur in both butt and top?
(16) Association of a given rot with other rots in same area of heartwood.
(17) Growth of rot mycelium in buried roots, humus, leaves, etc.
(18) Layers of mycelium in cheeks: leathery, chalky, incrusting, etc.
(19) Loss of weight in rotted wood.

Macroscopic characters of rot

(a) Early stage of rot, characteristics in radial, tangential, and cross-section views.
(b) Middle stage of rot, characteristics in radial, tangential, and cross-section views.

(c) Last stage of rot, characteristics in radial, tangential, and cross-section views.

Characters to be noted for each stage

(1) Changes in color of wood: color of rot, red, brown, white, etc.
(2) Changes in structure, as formation of holes, layers, strings, etc., or no marked changes in structure.
(3) Changes in spring wood or summer wood induced by rot.
(4) Consistency of the rot, as firm, soft, brittle, cheesy, etc.

Microscopic characters of rot

(1) Action of rot fungus on lignin, on cellulose.
(2) Action of rot fungus on cell contents, as on sugars, starches, etc., stored in wood.
(3) Action of rot fungus on medullary rays.
(4) Action of rot fungus on vessels or tracheids.
(5) Action of rot fungus on wood parenchyma.
(6) Action of rot fungus on wood fibers.
(7) Changes in structure and chemical nature of cell walls.
(8) Origin of changes, as from within outward or middle lamellae inward.
(9) Action on pits or torus of pits, on the three lamellae.
(10) Character of action on various wood elements, as marked destruction in structure visible under the microscope without staining or as shown only by using staining reagents.
(11) Holes, fissures, etc., in wood elements, due to enzymes or to action of fungus hyphae.
(12) Production of brown humus decomposition bodies and their deposit in cells.
(13) Enzymatic changes in wood in advance of fungus hyphae.
(14) How far, radially and vertically, the infecting hyphae extend beyond the visibly discolored incipient stage, determined by cultures and sectioning.
(15) Whether hyphae occur first in rays, vessels, wood parenchyma, or fibers.
(16) Along what wood elements do the hyphae travel the most rapidly? Is their movement radial or longitudinal? Do they move through the pits or bore directly through walls of the elements?
(17) Shape, size, color, location, and branching of hyphae; clamp connections and character of hyphae in the last stage of rot.
(18) Ultimate fate of each wood element for a given rot.
(19) Accumulation of carbon and ash constituents.

—W. H. LONG, Division of Forest Pathology, Bureau of Plant Industry, Albuquerque, New Mexico.

A disease of Hibiscus sabdariffa caused by Rhodochytrium.—The alga *Rhodochytrium Spilanthidis* Lagh. is known as a parasite of several wild-growing Angiosperms. Originally described by Lagerheim (1893) from *Spilanthes* (*Lundii*?) in Ecuador, it has subsequently been found to possess a wide host range, as well as a fairly extensive geographical distribution. Thus, it occurs in the Tropics and temperate zones of the New and the Old World on the following host plants.[1]

COMPOSITAE: *Ageratum conyzoides* L., *Ambrosia artemisiifolia* Bess., *A. trifida* L., *Solidago* sp., *Spilanthes acmella* Murray, *Sp.* (*Lundii*?), *Sp. Pseud-acmella* Hook & Arn.

ASCLEPIDACEAE: *Asclepias pumila* Vail.

The purpose of the present note is to record the occurrence of an identical Rhodochytrium on a number of the Malvaceae from which family the alga in question does not seem to have been reported. The new host plant is *Hibiscus sabdariffa* var. *altissima* Wester (the s. c. roselle), which is at present cultivated as a fiber plant here and there in the Tropics.

In an experimental planting of roselle on the east coast of Sumatra (Dutch East Indies) in 1926, it was noticed by the writer that a number of individuals—about 3 weeks old—showed a peculiar, stunted appearance. The bulk of the healthy plants had at that time reached an average height of 2 feet, while the stunted ones were about 1 foot high. Only the young internodes were shortened; the older ones were of normal length. The top leaves curled slightly downwards and, moreover, showed a pronounced distortion of the surface, indicating a retardation of growth of the main vein and of some of the veinlets. These parts were found to be thickly dotted on their lower side with small, bright red galls, containing the sporangia of a Rhodochytrium, which agreed in all respects with the current descriptions of *Rh. Spilanthidis* Lagh. Both resting spores and zoosporangia were found. A number of them also occurred on the leaf petioles and on the shortened internodes of the stem.

After some time the infected plants had resumed their normal growth above the infected region, which, however, remained as an indicator of the infection. The new growth proved to be free from Rhodochytrium, probably because a period of dry weather had set in, which prevented further upward spread of the parasites to the new growth. The diseased plants did not reach the same ultimate height as the healthy plants in the same field.

It deserves to be mentioned that Rhodochytrium generally does not cause appreciable deformation of its other known host plants. Some

[1] For further details see Palm, B. The geographical distribution of *Rhodochytrium*, in Ark. Bot. (Stockholm) **18**: No. 15. 1923.

species of Spilanthes sometimes do show, when infected, a somewhat abnormal elongation of internodes and petioles; in other host plants the only effect noticed is the production of the small galls. The action of Rhodochytrium on the roselle is thus the more remarkable.

As is well known, the sporangia of Rhodochytrium send haustoria-like prolongations into the fibrovascular bundles of the host tissue. The presence of the alga in these elements of a plant that, like roselle, is cultivated for its fibers, is thus clearly detrimental. So far, this disease is of no economic importance, having been found only in one field. Its occurrence should, however, be watched, as Rhodochytrium is widely spread in tropical and subtropical countries where the cultivation of roselle is most likely to be attempted.

No control measures have been necessary so far; should they become needed in the future, a scrupulously clean cultivation of the field for a short time after the germination of the seed would probably give sufficient protection. This was indicated by the fact that Rhodochytrium had infected the roselle from two of its weed host plants, *Spilanthes acmella* and *Ageratum conyzoides*, on which it occurred in great abundance in the experimental field mentioned.—B. T. PALM, Department of Botany, University of Illinois, Urbana, Ill.

A Chinese wheat resistant to flag smut.—Field studies of resistance in wheat to the flag smut organism, *Urocystis tritici* Koern., were initiated at the Agricultural Experiment Station, University of Nanking, China, in 1925, by Dr. R. H. Porter. Several Chinese wheat selections and a large number of foreign varieties were tested for their reaction to the flag-smut pathogen. These experiments have been continued up to the present time. The field experiments were first carried out by inoculating enough seed of each selection with smut spores to plant a 5-ft. row. Noninoculated seed was planted every 10th row as a check. After the first year the most susceptible selections were discarded, and the most promising resistant ones were inoculated and planted in rod rows repeated 5 to 10 times. As a result of these tests several selections of Chinese wheat were found to maintain their resistance consistently. One of these selections, now known as Nanking No. 716, has never shown any smut. It originated from a head selection collected by Dr. Porter at Weihsien, Shantung, in 1925. Because of its marked resistance, as well as its good growing habit, it was given a special supplementary greenhouse test in the fall of 1927 and again in 1928. The results of these tests, in each case, were read in the spring. The seeds were planted in 12-in. pots, filled with infected soil secured by mixing the soil for each pot with 50 gm. of smut spores. Five kernels of wheat were planted in each pot. The results are shown in the following table:

Name of wheat	1928			1929		
	Total number of plants	Number of infected plants	Percentage of infection	Total number of plants	Number of infected plants	Percentage of infection
Nanking 716	147	0.0	0.0	72	0.0	0.0
H. 1102	121	42.0	34.7	91	20.0	22.0
Nebawa	.....	.....	.....	65	0.0	0.0

Nabawa, a variety known to be resistant to flag smut in Australia, was secured from Waite Agricultural Research Institute, University of Adelaide, South Australia, and H. 1102 is a Chinese selection that has been used for checks because of its high susceptibility.

Nanking No. 716 will be increased and distributed for trial in various parts of China.—T. F. YU and H. K. CHEN, Plant Pathology Laboratory, University of Nanking, Nanking, China.

A machine for the treatment of small samples of seed grain with dust disinfectants.—In the course of the cereal-seed-disinfectant experiments at Madison, Wis., hundreds of different treatments are made for a single planting. The machine here described and illustrated speeds up the accurate and organized handling of such large numbers of small samples.

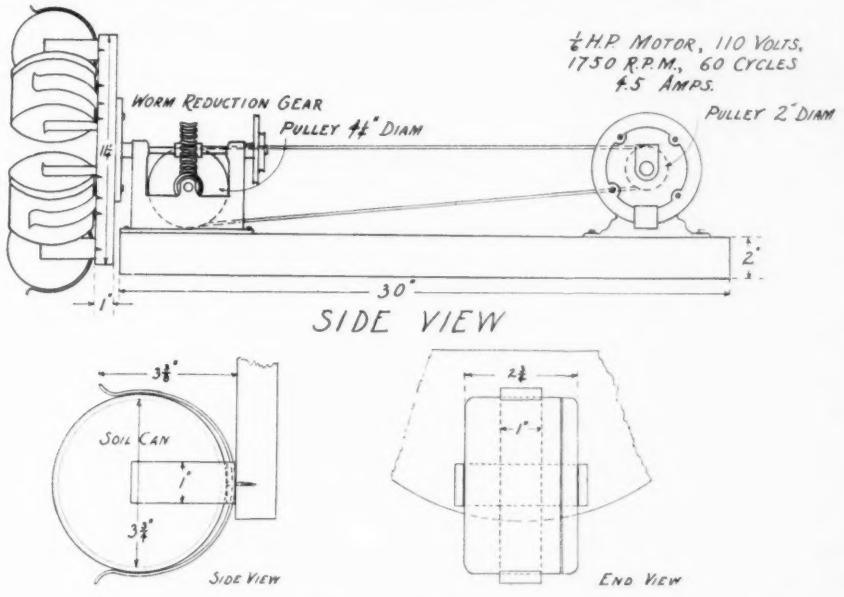

FIG. 1. Drawing showing dusting machine for treating small samples of grain.

This dusting machine (Fig. 1) consists of 6 soil-moisture cans clamped onto a revolving board by means of brass spring strips. The board is turned by a motor through the agency of a worm reduction gear, at the rate of 18 r. p. m., which speed is sufficiently slow to eliminate the cracking of grain and still fast enough so that 6 minutes' rotation thoroughly mixes a sample of grain with a dust disinfectant. Two sets of cans are used so that the operator can change and refill one set while the other set is revolving. When the seed is treated with excess dust and then screened before packeting one man can treat 60 samples of grain per hour, even when using a different disinfectant for each sample. When treating a previously weighed or measured series of samples, slightly more time is required for each treatment.

The cans filled ¾ full of grain hold 280 gms. of wheat, 215 gms. of barley, or 180 gms. of oats. When ¾ full the uniform mixing with the dust disinfectant is accomplished in a minimum of time.—W. H. THARP, Department of Plant Pathology, University of Wisconsin, Madison, Wis.

The prevention of arsenical injury to peach twigs and foliage in Virginia.—Peach twigs and foliage are very susceptible to arsenical injury and a great deal of damage has been caused in recent years in Virginia from lead-arsenate applications. Varieties vary considerably in their susceptibility to arsenical burning. Of the commercial varieties grown in Virginia, the J. H. Hale is the most susceptible; Elberta, next; and Belle of Georgia, the least susceptible.

Hydrated lime has been used extensively by peach growers to check this type of injury, but it has proved almost worthless as a preventive during wet seasons. This has made the lead-and-lime spray a very dangerous one, but the lack of anything better has compelled the growers to continue its use. A spray combination, however, that has recently proved very efficient in preventing arsenical injury to both twigs and foliage is the zinc-lime spray introduced by John W. Roberts, of the United States Department of Agriculture, for the control of the bacterial shot-hole disease. A 4–4–50 zinc-lime formula (zinc sulphate, 4 lbs.; hydrated lime, 4 lbs.; powdered lead arsenate, 1 lb., and water, 50 gals.) was used in approximately half of the commercial peach orchards in Virginia during the season of 1931, in all of the lead-arsenate applications. In every case where the zinc-lime material was used in all of the lead-arsenate applications scarcely a trace of arsenical burning could be found, whereas, in other orchards, where this material was not used, severe defoliation occurred, even if only one lead-arsenate application was made.

It is suggested that the zinc hydroxide, which is formed by chemical reaction between the hydrated lime and zinc sulphate, takes up the water-soluble arsenic in the form of an insoluble basic zinc arsenite.—R. H. HURT, Virginia Agricultural Experiment Station, Blacksburg, Virginia.

BOOK REVIEW

The Plant Rusts (Uredinales). By Joseph C. Arthur in collaboration with F. D. Kern, C. R. Orton, F. D. Fromme, H. S. Jackson, E. B. Mains, G. R. Bisby. iii–v + 446 pp., 186 figs. John Wiley & Sons, Inc., New York; Chapman & Hall, Limited, London, 1929. Price, $6.50.

Any discussion of the rusts by Dr. Arthur would be interesting because of the author's unique experience and eminence and because of the scientific standing of those who collaborated with him in writing the book. But "Plant Rusts" is interesting in its own right also because it contains a wealth of important information and expresses fundamental view-points.

In the first chapter is given an elementary but clear description of rusts in general. Chapter 2 summarizes in an interesting manner the historical development of uredinology. Chapter 3, on development and classification, plunges directly into details and concepts which presuppose a considerable previous knowledge on the part of the reader. The view-points expressed in this chapter seem refreshingly sound and sensible. For example, the statement on page 80 that the origin of rust species "can be explained more fully by tracing chromosomal inheritance" and the concept that heteroecism is merely one kind of specialization must appear to progressive mycologists as sensible. Such statements are gratifyingly modern, especially as there has been a tendency in the past to surround these phenomena with a somewhat thick veil of pseudoscientific mysticism. The directness of thought and reasoning is therefore noteworthy and commendable. Some mycologists may be inclined, however, to question the statements regarding the degrading effect of parasitism, as, for example, the following statement on page 97: "The mode of life has disturbed and exaggerated the balance between vegetative and reproductive activities greatly in favor of the latter as compared with non-parasitic plants." Whether the mode of life really is responsible for the development of peculiarities in the rusts may be open to question.

In the discussion of classification it is frankly admitted that the classification of rusts attempts to meet practical requirements and does not necessarily indicate natural affinities, desirable as the latter might be. The position with respect to *nomina conservanda* will probably be welcomed by most disciples of common sense, even in mycology.

The cytological and morphological features of the rusts are well summarized and illustrated in chapter 4, although the significance of cytological phenomena is given relatively little consideration. Possibly it is as well: in the past there often has been too much speculation regarding the meaning, real or fancied, of certain phenomena.

The dissemination and geographic distribution, especially the latter (Chapter 5), are well and adequately discussed. The chapter is packed with interesting facts, and the interpretations and conclusions seem sound. In chapter 6 the phenomena of spore germination and the factors affecting germination, the subsequent development of germ tubes, their entrance into the host, and the development of mycelium and haustoria are described for a number of rusts. There is a brief discussion also of the effect of environmental factors on the development of rusts. Some pathologists may wish to improve the definition of infection given on page 231, but the term has no precisely standardized meaning, and the author of the book has at least made his concept fairly clear. A fuller discussion of the effect of environmental factors on infection and the subsequent development of the rust might have been desirable, but perhaps this would have exceeded the scope intended for the book.

Chapter 7 is devoted to a consideration of physiologic specialization and the nature of resistance and susceptibility. The section on specialization probably could have been made clearer by a more liberal use of keys and tables. The writer would be inclined to take issue with some of the statements in the discussion of resistance, but, as they are matters of opinion and interpretation, there probably is no need to go into details.

Chapter 8 contains interesting information on abnormalities and diseases of rusts, together with a description of pathologic changes caused in host plants.

Chapter 9 is devoted to a discussion of economic considerations. One obtains a good idea of the importance of the most destructive rusts of the principal groups of economic plants, although the discussions are necessarily brief. The last chapter, on methods of investigation, brings together some useful information on the collection and preservation of specimens, the technic for microscopic study, and culture methods. The index leaves something to be desired, and it is hoped that it will be amplified in subsequent editions.

The sequence of subjects discussed in the various chapters is logical, and, despite the difficulties pointed out in the preface to the book, a rather high degree of unity has been attained. The style is clear, for the most part, and, except in a few places, the statements are direct and free from ambiguities. There is some repetition, but this probably is not only unavoidable but even desirable.

The desirability of uniform terminology in mycology becomes evident in reading the book. The terms sporophyte and gametophyte are used in the book in preference to the terms haplophase, dikaryophase, and diplophase. The writer suggests that the latter are more precise and more nearly meet the requirements, especially in the rust and smut fungi. The use of

the terms physiologic race and physiologic form may lead to further confusion where much already exists. The use of "physiologic race" to denote the physiological entities such as *Puccinia graminis tritici*, while accepting the term physiologic form for the component entities, partly disregards previous usage without clarifying the situation. The statements regarding specialization are not always clear because, in at least some instances, terms are used loosely or inconsistently. For example, on page 260 one reads: "Similar situations occur in other weakly specialized rusts such as *P. graminis* and *P. coronata.*" But on page 336 the following statement is made: ". . . the species (*P. coronata*) does not show the sharp host specialization or separation into races which is shown by *P. graminis.*" Specialization probably is considered from different viewpoints in the two statements, and the result is an apparent inconsistency. The propriety of designating as "mutable species" the long-cycle rusts "which pass readily at times from the long-cycle to a condition that actually is, or else resembles a short-cycle condition" may also be questioned by those who would restrict the meaning of the word mutable to a tendency to change genotypically. These few minor defects, if they are defects, do not detract appreciably, however, from the general excellence of the book.

The voluminous literature on rusts has been well and fairly summarized. In a few instances reference is not made to papers in which important phenomena were first described; instead, later and more comprehensive papers are cited. The reader in such instances does not get the historical perspective quickly. The problem of selecting literature for citation is, of course, a difficult one, and the relative importance of contributions is partly a matter of personal opinion. It would have been natural to feature the extensive investigations of the author and his associates. This tendency, however, has been avoided and the material is presented with a high degree of objectivity.

The scientific tone of the book is excellent throughout. Controversial questions are discussed with dignity, fairness, and excellent judgment. There is a nice balance between fact and philosophy, with a soundness of viewpoint that would, of course, be expected of a man of the author's eminence and the high scientific attainments of his associates, but which nevertheless does them credit.

Botanists in general and mycologists in particular owe Dr. Arthur and his associates a debt of gratitude for bringing together in book form so many facts regarding rusts and, especially, the rich experience and broad viewpoints obtained from their own extensive and fruitful investigations.—E. C. STAKMAN, University Farm, St. Paul, Minnesota.

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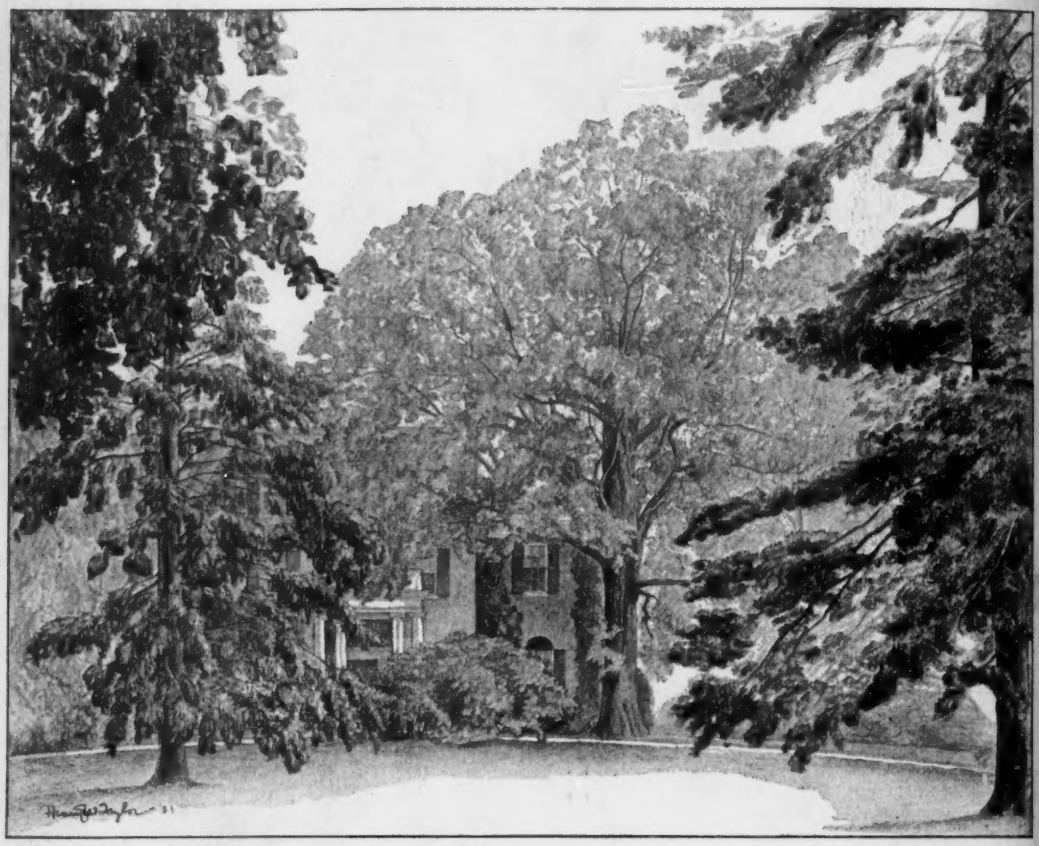

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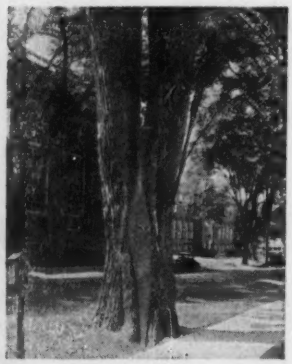

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